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Dialog level 05.27.00D

Last logoff: 13oct09 15:07:59

Logon file405 19oct09 07:47:50

*** ANNOUNCEMENTS ***

*** FREE FILE OF THE MONTH: OCTOBER
American Business Directory (File 531)

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EMBASE Classic (File 772) available to all customers.

NEW FILE

***File 558, Mergent China Private Company Database

***File 457, The Lancet(R)

FILE RENAMED

***File 323, RAPRA: Rubber & Plastics is now RAPRA Polymer Technology

RESUMED UPDATING

***File 523, D&B European Financial Records

RELOADS COMPLETED
***File 663, TRADEMARKSCAN(R) - Monaco
***File 676, TRADEMARKSCAN(R) - Slovak Republic
***File 677, TRADEMARKSCAN(R) - Liechtenstein
***File 681, TRADEMARKSCAN(R) - Hungary
***File 683, TRADEMARKSCAN(R) - Ireland
***File 685, TRADEMARKSCAN(R) - Lithuania
***File 688, TRADEMARKSCAN(R) - Portugal
***File 697, TRADEMARKSCAN(R) - Latvia

FILES REMOVED

***File 743, New Jersey, The Record - Please use NewsRoom
***File 301, CHEMNAME - Please use File 398 ChemSearch
***File 388, PEDS: Defense Program Summaries
***File 588, DMS-FI Contract Awards
***File 559, CorpTech Directory of Technology Cos.

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>>>a specific database by entering HELP NEWS <file number>. <<<
* * *

SYSTEM:HOME

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Menu System II: D2 version 1.8.0 term=ASCII

*** DIALOG HOMEBASE(SM) Main Menu ***

Information:

1. Announcements (new files, reloads, etc.)
2. Database, Rates, & Command Descriptions
3. Help in Choosing Databases for Your Topic
4. Customer Services (telephone assistance, training, seminars, etc.)
5. Product Descriptions

Connections:

6. DIALOG(R) Document Delivery
7. Data Star(R)

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/H = Help

/L = Logoff

/NOMENU = Command Mode

Enter an option number to view information or to connect to an online service. Enter a BEGIN command plus a file number to search a database

(e.g., B1 for ERIC).

? b 410

19oct09 07:47:51 User226352 Session D1178.1
\$0.00 0.267 DialUnits FileHomeBase
\$0.00 Estimated cost FileHomeBase
\$0.00 Estimated cost this search
\$0.00 Estimated total session cost 0.267 DialUnits

File 410:The Chronolog 2009
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Set Items Description
--- ---
? set hi ;set hi
HIGHLIGHT set on as ''
HIGHLIGHT set on as ''
? b biochem

19oct09 07:47:57 User226352 Session D1178.2
\$0.00 0.115 DialUnits File410
\$0.00 Estimated cost File410
\$0.02 TELNET
\$0.02 Estimated cost this search
\$0.02 Estimated total session cost 0.382 DialUnits

SYSTEM:OS - DIALOG OneSearch

File 5:Biosis Previews(R) 1926-2009/Oct W2
(c) 2009 The Thomson Corporation
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(c) 2009 NTIS, Intl Cpyrght All Rights Res
File 24:CSA Life Sciences Abstracts 1966-2009/Nov
(c) 2009 CSA
File 34:SciSearch(R) Cited Ref Sci 1990-2009/Oct W2
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File 40:Enviroline(R) 1975-2008/May
(c) 2008 Congressional Information Service

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File 41:Pollution Abstracts 1966-2009/Nov
(c) 2009 CSA
File 45:EMCare 2009/Oct W2
(c) 2009 Elsevier B.V.
File 50:CAB Abstracts 1972-2009/Oct W2
(c) 2009 CAB International
File 65:Inside Conferences 1993-2009/Oct 16
(c) 2009 BLDSC all rts. reserv.
File 71:ELSEVIER BIOBASE 1994-2009/Oct W3
(c) 2009 Elsevier B.V.

*File 71: The file has been reloaded. Accession numbers have changed.

File 72:EMBASE 1993-2009/Oct 19
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*File 72: The file has been synchronized to the calendar date. It is complete and up to date as of 9/28/2009.

File 73:EMBASE 1974-2009/Oct 19

(c) 2009 Elsevier B.V.
*File 73: The file has been synchronized with the calendar date.
It is complete and up to date as of 9/28/2009.
File 76:Environmental Sciences 1966-2009/Nov
(c) 2009 CSA.
File 98:General Sci Abs 1984-2009/Oct
(c) 2009 The HW Wilson Co.
File 103:Energy SciTec 1974-2009/Oct B1
(c) 2009 Contains copyrighted material
*File 103: For access restrictions see Help Restrict.
File 136:BioEngineering Abstracts 1966-2007/Jan
(c) 2007 CSA.
*File 136: This file is closed.
File 143:Biol. & Agric. Index 1983-2009/Sep
(c) 2009 The HW Wilson Co
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(c) 2009 INIST/CNRS
File 154:MEDLINE(R) 1990-2009/Oct 16
(c) format only 2009 Dialog
File 155:MEDLINE(R) 1950-2009/Oct 16
(c) format only 2009 Dialog
File 156:ToxFile 1965-2009/Oct W2
(c) format only 2009 Dialog
File 162:Global Health 1983-2009/Oct W2
(c) 2009 CAB International
File 172:EMBASE Alert 2009/Oct 19
(c) 2009 Elsevier B.V.
*File 172: The file has been synchronized with today's calendar date.
It
is complete and up to date.
File 305:Analytical Abstracts 1980-2009/Aug W4
(c) 2009 Royal Soc Chemistry
*File 305: Alert feature enhanced for multiple files, duplicate
removal, customized scheduling. See HELP ALERT.
File 369:New Scientist 1994-2009/Oct W2
(c) 2009 Reed Business Information Ltd.
File 370:Science 1996-1999/Jul W3
(c) 1999 AAAS
*File 370: This file is closed (no updates). Use File 47 for more
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information.
File 393:Beilstein Database - Abstracts 2008/Q2
(c) 2008 Beilstein GmbH
File 399:CA SEARCH(R) 1967-2009/UD=15116
(c) 2009 American Chemical Society
*File 399: Use is subject to the terms of your user/customer
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IPCR/8 classification codes now searchable as IC=. See HELP NEWSIPCR.
File 434:SciSearch(R) Cited Ref Sci 1974-1989/Dec
(c) 2006 The Thomson Corp

Set Items Description

```
--- -----
? s amazonensis and promastigote and surface and antigen?
Processing
Processed 20 of 29 files ...
Completed processing all files
    12167 AMAZONENSIS
    14701 PROMASTIGOTE
    8592688 SURFACE
    4820606 ANTIGEN?
S1      164 AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND
ANTIGEN?
? rd s1

>>>Duplicate detection is not supported for File 393.

>>>Records from unsupported files will be retained in the RD set.
S2      51 RD S1 (unique items)
? s amazonensis and ((promastigote and surface and antigen?) or PSA
or ESA)
Processing
Processed 20 of 29 files ...
Completed processing all files
    12167 AMAZONENSIS
    14701 PROMASTIGOTE
    8592688 SURFACE
    4820606 ANTIGEN?
    136324 PSA
    40318 ESA
S3      175 AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND
ANTIGEN?) OR PSA OR ESA)
? rd s3

>>>Duplicate detection is not supported for File 393.

>>>Records from unsupported files will be retained in the RD set.
S4      55 RD S3 (unique items)
? ds

Set      Items   Description
S1      164 AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND ANTIGEN?
S2      51 RD S1 (unique items)
S3      175 AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND
ANTIGEN?) OR
                           PSA OR ESA)
S4      55 RD S3 (unique items)
? s ((promastigote and surface and antigen?) or PSA or ESA)
Processing
Processed 20 of 29 files ...
Completed processing all files
    14701 PROMASTIGOTE
    8592688 SURFACE
```

```

4820606  ANTIGEN?
136324  PSA
40318  ESA
S5  177478 ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)
? s s5 and (leishmania or amazonensis)
177478 S5
159840 LEISHMANIA
12167 AMAZONENSIS
S6  1112 S5 AND (LEISHMANIA OR AMAZONENSIS)
? rd s6

>>>Duplicate detection is not supported for File 393.

>>>Records from unsupported files will be retained in the RD set.
S7  335 RD S6 (unique items)
? s s7 nor PY>2004
>>>Term "NOR" in invalid position
? s s7 not PY>2004
Processing
Processed 10 of 29 files ...
Completed processing all files
335 S7
37837515 PY>2004
S8  298 S7 NOT PY>2004
? ds

Set      Items    Description
S1      164      AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND ANTIGEN?
S2      51       RD S1 (unique items)
S3      175      AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND
ANTIGEN?) OR
                  PSA OR ESA)
S4      55       RD S3 (unique items)
S5  177478 ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)
S6  1112 S5 AND (LEISHMANIA OR AMAZONENSIS)
S7  335 RD S6 (unique items)
S8  298 S7 NOT PY>2004
? s s8 and 42.5
298 S8
0 42.5
S9  0 S8 AND 42.5
? s s8 and kda
298 S8
958548 KDA
S10 52 S8 AND KDA
? t s10/7/all
>>>Format 7 is not valid in file 143

10/7/1      (Item 1 from file: 5)
DIALOG(R)File  5:Biosis Previews(R)

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17674704 BIOSIS NO.: 200400045461

Immunolocalization of Leishmania (Viannia) braziliensis membrane antigens recognized by mAbs SST-2, SST-3, and SST-4.

AUTHOR: Silveira T G V; Takahashi H K; Straus A H (Reprint)

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Paulo/Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP,
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JOURNAL: Parasitology 127 (5): p449-456 November 2003 2003

MEDIUM: print

ISSN: 0031-1820

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: The immunolocalization of Leishmania (Viannia) braziliensis stage-specific antigens recognized by mAbs was analysed by transmission electron microscopy. The antigen recognized by mAb SST-2 was present at the surface of promastigotes, including the flagellum and flagellar pocket. The reactivity of SST-2 with isolates of different serodemes showed a pronounced microheterogeneity in terms of the number of reactive bands within the low molecular weight range from 24 to 33 kDa. The 180 kDa glycoprotein recognized by mAb SST-3 was present only in the flagellar membrane. SST-3 also recognized multiple discrete bands from 160 to 200 kDa, as observed in several serodemes. In contrast, mAb SST-4, which recognizes a 98 kDa antigen, showed weak labelling on the promastigote surface by transmission electron microscopy and indirect immunofluorescence. Based on Western blotting, indirect immunofluorescence, and solid-phase radioimmunoassay, the antigens recognized by mAbs SST-2, SST-3 and SST-4 were present in all L. (V.) braziliensis analysed, from 7 different serodemes.

10/7/2 (Item 2 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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17657659 BIOSIS NO.: 200400028416

Immune complex antigens as a tool in serodiagnosis of kala-azar.

AUTHOR: Chakraborti Tapati (Reprint); Sarkar Dwijen; Ghosh Dilip K

AUTHOR ADDRESS: Department of Biochemistry and Biophysics, University of

Kalyani, Kalyani, WB, 741235, India**India

AUTHOR E-MAIL ADDRESS: tchakraborti@yahoo.com
JOURNAL: Molecular and Cellular Biochemistry 253 (1-2): p191-198
November
2003 2003
MEDIUM: print
ISSN: 0300-8177 _ (ISSN print)
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The 63 kDa surface antigen of Leishmania promastigotes is one of the most important virulent factors in establishing the host parasite relationship. This glycoprotein is revealed by surface iodination study as well as by metabolic labeling and immunoblot methods. In search of this specific antigen for serodiagnosis, immune complexes (ICs) were isolated from kala-azar

patient sera and analysed by SDS-PAGE and Western immunoblotting. The

immunoblot of kala-azar IC with patient sera, anti-promastigote sera and anti gp63 sera detected the major antigen of 55 kDa. This recognition suggests that 55 kDa antigen and gp63 have common antigenic epitope(s). Normal IC did not react with anti gp63 sera indicating absence of this antigen in normal IC. To confirm the parasitic origin of the 55 kDa antigen of kala-azar IC, in vitro IC was formed with parasite antigen and acid dissociated kala-azar IC antibody. This indicated the antigenic similarity of the 55 kDa antigen and gp63 antigen of the parasite. This also suggested that the former antigen may have been processed from gp63. In summary, identification of parasite antigen (55 kDa) in IC of kala-azar patients' sera may be useful in developing a serodiagnostic assay of visceral leishmaniasis. Several other antigens are visualized in kala-azar IC when developed with patient sera. But specificity and efficacy of these antigens have not yet been evaluated in serodiagnosis of the disease.

10/7/3 (Item 3 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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17626210 BIOSIS NO.: 200300576887
gp63 homologues in Trypanosoma cruzi: Surface antigens with metalloprotease activity and a possible role in host cell infection.
AUTHOR: Cuevas Ileana C; Cazzulo Juan J; Sanchez Daniel O (Reprint)
AUTHOR ADDRESS: Instituto de Investigaciones Biotecnologicas,
Universidad Nacional de General San Martin, INTI, Edificio 24, 1650, San Martin,
Buenos Aires, Argentina**Argentina
AUTHOR E-MAIL ADDRESS: dsanchez@iib.unsam.edu.ar
JOURNAL: Infection and Immunity 71 (10): p5739-5749 October 2003 2003
MEDIUM: print

ISSN: 0019-9567 _(ISSN print)

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: gp63 is a highly abundant glycosylphosphatidylinositol (GPI)-anchored membrane protein expressed predominantly in the promastigote but also in the amastigote stage of *Leishmania* species. In *Leishmania* spp., gp63 has been implicated in a number of steps in establishment of infection. Here we demonstrate that *Trypanosoma cruzi*, the etiological agent of Chagas' disease, has a family

of gp63 genes composed of multiple groups. Two of these groups,

Tcgp63-I

and -II, are present as high-copy-number genes. The genomic organization

and mRNA expression pattern were specific for each group. Tcgp63-I was

widely expressed, while the Tcgp63-II group was scarcely detected in Northern blots, even though it is well represented in the *T. cruzi* genome. Western blots using sera directed against a synthetic peptide

indicated that the Tcgp63-I group produced proteins of apprx78 kDa, differentially expressed during the life cycle. Immunofluorescence staining and phosphatidylinositol-specific phospholipase C digestion confirmed that Tcgp63-I group members are surface proteins bound to the membrane by a GPI anchor. We also demonstrate the presence of metalloprotease activity which is attributable, at least in part, to Tcgp63-I group. Since antibodies against Tcgp63-I partially blocked infection of Vero cells by trypomastigotes, a possible role for this group in infection is suggested.

10/7/4 (Item 4 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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17611265 BIOSIS NO.: 200300579984

Surface glycoprotein PSA (GP46) expression during short- and long-term culture of *Leishmania chagasi*.

AUTHOR: Beetham Jeffrey K (Reprint); Donelson John E; Dahlin Rebecca R

AUTHOR ADDRESS: Department of Veterinary Pathology, Iowa State University,

2714 Veterinary Medicine, Ames, IA, 50011, USA**USA

AUTHOR E-MAIL ADDRESS: jbeetham@iastate.edu

JOURNAL: Molecular & Biochemical Parasitology 131 (2): p109-117 October

2003 2003

MEDIUM: print

ISSN: 0166-6851

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: The mRNAs encoding promastigote surface antigen (PSA) of *Leishmania chagasi* have previously been shown to increase about 30-fold as in vitro cultured parasites progress from logarithmic to stationary phase, growth phases that are, respectively

associated with parasites having low and high infectivity to mammals.

Experiments reported here establish by western blot analysis that PSA proteins of 44 and 66 kDa also increase about 30-fold as parasite cultures reach stationary phase. Serial passage of parasite cultures resulted in a progressive reduction in PSA protein and RNA abundance to levels less than 3% that of cultures newly-initiated with

parasites derived from a parasitized rodent. Loss of PSA mRNA abundance in serially passaged cells was not due to reduced PSA gene transcription rates, as determined by nuclear run-on assays.

Neither

was the loss associated with a marked decrease in PSA mRNA stability. Analysis of PSA RNA stability in the presence of actinomycin D, an inhibitor of transcription elongation, failed to detect

a difference in fully processed cytosolic PSA mRNA stability regardless of the number of times a culture was passaged or the growth phase of the culture. Based on the lack of detectable difference in (cytosolic) mature PSA mRNA stability during promastigote development, the data indirectly suggest that the regulated expression of

PSA in cells from low-passage cultures and the loss of PSA expression in high-passage cultures may be mediated by nuclear events

that occur after transcription of the PSA genes and before arrival of the mature mRNAs in the cytoplasm.

10/7/5 (Item 5 from file: 5)
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16745135 BIOSIS NO.: 200200338646

Comparison of the post-transcriptional regulation of the mRNAs for the surface proteins PSA (GP46) and MSP (GP63) of *Leishmania chagasi*

AUTHOR: Myung Karen S; Beetham Jeffrey K; Wilson Mary E; Donelson John E

(Reprint)

AUTHOR ADDRESS: Department of Biochemistry, University of Iowa, Iowa City,

IA, 52242, USA**USA

JOURNAL: Journal of Biological Chemistry 277 (19): p16489-16497 May 10,

2002 2002

MEDIUM: print

ISSN: 0021-9258

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: MSP (GP63) and PSA (GP46) are abundant 63- and 46-kDa glycolipid-anchored proteins on the surface of the promastigote form of

most Leishmania species. MSP is a zinc metalloprotease that confers resistance to host complement-mediated lysis. PSA contains internal repeats of 24 amino acids, and its function is unknown. The steady state

levels of mRNAs for both glycoproteins are regulated post-transcriptionally, resulting in about a 30-fold increase as Leishmania chagasi promastigotes grow in vitro from logarithmic phase to stationary phase. Previous studies showed the

3'-untranslated regions (3'-UTRs) of these mRNAs are essential for this post-transcriptional regulation. These two 3'-UTRs of 1.0 and 1.3 kilobases were cloned immediately down-stream of a beta-galactosidase

reporter gene in a plasmid, and segments were systematically deleted to

examine which portions of the 3'-UTRs contribute to the post-transcriptional regulation. The 92-nucleotide segment of greatest

similarity between the two 3'-UTRs was deleted without loss of regulation, but the segments flanking this similarity region have positive regulatory elements essential for the regulation. We propose

that similar, but non-identical, molecular mechanisms regulate the parallel expression of these two *L. chagasi* mRNAs despite their lack of

sequence identity. These post-transcriptional mechanisms resemble the mechanism recently suggested for the regulation of mRNAs encoding the

dipeptide (EP) and pentapeptide (GPEET) repeat proteins in Trypanosoma

brucei that involves interactions between positive and negative regulatory elements in the 3'-UTR.

10/7/6 (Item 6 from file: 5)
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16383340 BIOSIS NO.: 200100555179

Inhibition of macrophage invasion by monoclonal antibodies specific to Leishmania (Viannia) braziliensis promastigotes and

characterisation of their antigens

AUTHOR: Silveira Thais G V; Suzuki Erika; Takahashi Helio K; Straus Anita H

(Reprint)

AUTHOR ADDRESS: Department of Biochemistry, Universidade Federal de Sao

Paulo/Escola Paulista de Medicina, Rua Botucatu 862, Sao Paulo, SP,
04023-900, Brazil**Brazil

JOURNAL: International Journal for Parasitology 31 (13): p1451-1458
November, 2001 2001

MEDIUM: print

ISSN: 0020-7519

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: Monoclonal antibodies that specifically recognise *Leishmania (Viannia) braziliensis* promastigotes were produced and termed SST-2, SST-3 and SST-4. SST-2 recognises a conformational epitope present in a 24-28 kDa doublet and in a 72 kDa component, as verified by Western blotting. Indirect immunofluorescence showed that the

antigen recognised by SST-2 is distributed homogeneously on the parasite surface. SST-3 recognises a flagellar glycoprotein of apprx180 kDa. The reactivity of this mAb was abolished by sodium m-periodate treatment, indicating that SST-3 reacts with a carbohydrate

epitope of the 180 kDa antigen. SST-4 recognises a conformational epitope of a 98 kDa antigen. SST-2, SST-3 and SST-4 were specific to *L. (V.) braziliensis* promastigote forms. Indirect immunofluorescence did not show reactivity of SST-2 or

SST-3

with amastigotes of *L. (V.) braziliensis*, or with promastigotes of *Leishmania (Viannia) panamensis*, *Leishmania (Viannia) guyanensis*, *Leishmania (Viannia) naiffi*, *Leishmania (Viannia) lainsoni*, *Leishmania (Leishmania) amazonensis*, *Leishmania (Leishmania) major*, or *Leishmania (Leishmania) chagasi*. We also evaluated the involvement of SST-2, SST-3 and SST-4 antigens in parasite-macrophage interaction. Fab fragments of SST-3 and SST-4 significantly inhibited the infectivity of

L. (V.) braziliensis promastigotes to mouse peritoneal macrophages.

10/7/7 (Item 7 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

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14658075 BIOSIS NO.: 199800452322

Leishmania major: Cell type dependent distribution of a 43 kDa antigen related to silent information regulatory-2 protein family

AUTHOR: Zemzoumi Khalid; Sereno Denis; Francois Celine; Guilvard Elaine;
Lemesre Jean-Loup; Ouaissi Ali (Reprint)
AUTHOR ADDRESS: CJF INSERM 96-04, Centre ORSTOM Montpellier, 911 Ave.
Agropolis, BP 5045, 34032 Montpellier Cedex 01, France**France
JOURNAL: Biology of the Cell (Paris) 90 (3): p239-245 June, 1998 1998
MEDIUM: print
ISSN: 0248-4900
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: In previous studies we have characterized several Leishmania major polypeptides and showed that one member of this group (LmSIR2rp) shared significant homology to silent information regulator 2 (SIR2) of *Saccharomyces cerevisiae*, a protein playing a role in both telomeric and mating type loci repression in these organisms. In the present study, by using molecular and immunological approaches, we could identify LmSIR2rp homologues in different Leishmania species and developmental stages (eg logarithmic (LP) and stationary phase promastigotes (SP) and amastigotes). The reactive antigen was also detected in *Trypanosoma cruzi* extracts. Surprisingly, immunofluorescence assays revealed that LmSIR2rp is associated mainly with cytoplasmic granules of different sizes and numbers depending on the life stage of the parasite used. No reactivity was observed in the nucleus, in agreement with the Western blot showing an absence of immunoreactivity of anti-LmSIR2rp immune serum against parasite nuclear extracts. Furthermore, immunoprecipitation of (35S)methionine-labeled promastigote antigens after pulse chase experiments, using anti-LmSIR2rp fusion protein antibodies, showed that the protein is among parasite excreted-secreted antigens (ESA). Moreover, immunofluorescence assays conducted with short time incubations of either purified LmSIR2rp or viable promastigotes with murine macrophages, revealed that LmSIR2rp could be bound to the macrophage surface. The unexpected cytoplasmic localization of LmSIR2rp and its presence in ESA may suggest a new mode of action for silent information regulatory factor homologues.

13031893 BIOSIS NO.: 199598499726
Expression of antigens in virulent and avirulent Indian strains of
Leishmania donovani
AUTHOR: Sanyal Tapati; Gangopadhyay Prosenjit; Ghosh Dilip K; Sarkar
Dwijen
(Reprint)
AUTHOR ADDRESS: Dep. Cell Biology, Indian Inst. Chemical Biology, 4
Raja S.
C. Mullick Road, Calcutta 700 032, India**India
JOURNAL: Journal of Biosciences (Bangalore) 19 (3): p291-299 1994
1994
ISSN: 0250-5991
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Indo-Gen mediated surface labelling with ^{125}I demonstrated differences in surface oriented antigens between virulent and avirulent promastigotes of Leishmania donovani. In case of virulent strains, surface polypeptides with molecular masses of 63, 53, 42 and 38 kDa were found to be labelled with ^{125}I whereas in the case of avirulent stains 68, 55, 50, 46, 42 and 33 kDa components were iodinated. Further studies by immunoblot assay using different subcellular fractions of virulent and avirulent parasites demonstrated that antibody raised against gp63 cross-reacted with the 63 and 60 kDa antigen of the virulent and avirulent Leishmania donovani strains of Indian origin respectively. It indicates that these two polypeptides are antigenically similar. When virulent and avirulent cells were grown in the presence of varying concentration of tunicamycin and immunoblotted with anti gp63, it was observed that with increasing concentration of tunicamycin the 63 kDa polypeptide of the virulent cells shifted to approximately 58-57 kDa and the 60 kDa polypeptide of the avirulent cells shifted to 57 kDa. This suggests that glycosylation may play an important role in antigenic variation between virulent and avirulent parasites.

10/7/9 (Item 9 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)
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12817543 BIOSIS NO.: 199598285376
Detection of serum antibodies against Leishmania 94 kDa antigen in visceral and cutaneous leishmaniosis due to Leishmania infantum
AUTHOR: Rolland L (Reprint); Belkaid M; Seye A; Schneider P; Gentilini M
AUTHOR ADDRESS: 1 rue Lamartine, F-38000 Grenoble, France**France

JOURNAL: Parasite 2 (1): p13-21 1995 1995
ISSN: 1252-607X
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Leishmania promastigotes polypeptides are analyzed by immunoblotting with sera from patients infected with different Leishmania species and presenting visceral or cutaneous infections. These sera recognize Leishmania polypeptides in several molecular masses. The major findings of this study are as follow. 1) The Leishmania 94 kDa antigen, which is specifically recognized by all sera from *L. infantum*-infected patients with visceral infection, is recognized by some sera from *L. infantum*-infected patients presenting cutaneous infection. 2) All patients with cutaneous infections due to *L. tropica*, *L. amazonensis*, or *L. guyanensis* do not develop anti-94 kDa antibodies, whatever the Leishmania species used as antigens. 3) Difference in electrophoretic mobilities is seen between the 94 kDa antigen identified by sera from Leishmania infantum-infected patients, and the antigen both recognized by the Concanavalin A lectin and a rabbit antiserum raised against deglycosylated Promastigote Surface Protease.

10/7/10 (Item 10 from file: 5)
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12542091 BIOSIS NO.: 199598009924
The Kinetoplastid Membrane Protein 11 of Leishmania donovani and African Trypanosomes Is a Potent Stimulator of T-Lymphocyte Proliferation
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JOURNAL: Infection and Immunity 62 (11): p4893-4899 1994 1994
ISSN: 0019-9567
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Kinetoplastid membrane protein 11 (KMP-11) from Leishmania donovani is an abundant 11-kDa surface membrane glycoprotein. Lymph node cells from mice of six different H-2 haplotypes immunized with

KMP-11 or with *L. donovani* promastigotes were stimulated to proliferate *in vitro* with purified KMP-11. Primed purified T cells required antigen presentation since they were not stimulated unless KMP-11-pulsed or *L. donovani*-infected macrophages were added. Promastigotes of a wide variety of *Leishmania* species and procyclic forms of African trypanosomes stimulated proliferation of KMP-11-primed or *L. donovani* promastigote-primed lymph node cells. All of the *Leishmania* promastigotes and African trypanosomes tested contained an 11-kDa protein, as detected by immunoblotting with KMP-11-specific monoclonal antibodies. The widespread distribution of the 11-kDa (KMP-11) molecules and their ability to stimulate strong T-lymphocyte proliferation in a non-H-restricted fashion suggest that they may be important molecules for induction of cell-mediated immune responses.

10/7/11 (Item 11 from file: 5)
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12069197 BIOSIS NO.: 199497090482
Relationships between cell surface protease and acid phosphatase activities of *Leishmania* promastigote
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JOURNAL: Molecular Biology Reports 18 (3): p189-195 1993 1993
ISSN: 0301-4851
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: A correlation between the ratio of the cell surface protease activity to phosphatase activity and the complexity of the pattern of cell surface exposed polypeptides of *Leishmania* promastigotes was demonstrated for various strains grown under similar conditions. The ratio of the cell surface protease activity to acid phosphatase activity was high for *L. major* and *L.b. panamensis* and it correlates with the expression of a single polypeptide of 63 KDa on their cell surface. Intermediate and lower ratios of these enzymatic activites relate with more complex radioiodinated patterns: two

main bands in *L.b. guyanensis* (70 and 58 KDa) and *L.b. braziliensis* (72 and 60 KDa) and three main bands 65, 50, 27 KDa in all *L.m. mexicana* strains tested. Evidence is presented that the acid phosphatase located on the *L.m. mexicana* cell surface is not an artifact due to a secondary absorption of the secreted acid phosphatase from the culture medium. These results confirm the *Leishmania* antigen cell surface heterogeneity. The implications on the biology of *Leishmania* and the clinical manifestation of leishmaniasis are discussed.

10/7/12 (Item 12 from file: 5)
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11972681 BIOSIS NO.: 199396137097
Antigen-reactive gamma-delta T cells in human leishmaniasis
AUTHOR: Russo Donna M; Armitage Richard J; Barral-Netto Manoel; Barral Aldina; Grabstein Kenneth H; Reed Steven G (Reprint)
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JOURNAL: Journal of Immunology 151 (7): p3712-3718 1993
ISSN: 0022-1767
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: The importance of Ag-specific gamma-delta T lymphocytes in human immune responses to pathogenic organisms is unknown. In the present study the expression of gamma-delta TCR on T lymphocytes from patients with cutaneous, mucosal, or visceral leishmaniasis was examined. All of these patient groups had elevated levels of gamma-delta T cells in peripheral blood. Patients' gamma-delta T cells included CD8+ as well as null cells. The percentage of T cells expressing gamma-delta TCR was increased significantly by stimulation in vitro with certain parasite Ag. T-cell lines generated by stimulation with promastigote lysates of *Leishmania amazonensis* or *L. braziliensis* typically contained 25 to 60% gamma-delta T cells. In contrast, two immunodominant surface Ag of *L. amazonensis*, gp63 and gp42, did not expand gamma-delta T cells from infected patients, although both Ag elicited strong alpha-beta T-cell responses. gamma-delta T cells isolated from a

Leishmania-specific T-cell line responded to stimulation with promastigote lysate. Of particular interest, gamma-delta T cells from PBMC of a patient with mucosal leishmaniasis responded to stimulation with a recombinant 70 kDa heat shock protein of L. chagasi. This study demonstrated that several clinical forms of leishmaniasis induced elevated numbers of gamma-delta T cells that responded specifically to Leishmania Ag in vitro. Therefore, this component of the T-cell response to Leishmania may impact the outcome of clinical disease.

10/7/13 (Item 13 from file: 5)
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11841168 BIOSIS NO.: 199396005584
Leishmania donovani surface glycoconjugate GP36 is the major immunogen component of the fucose-mannose ligand (FML)
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JOURNAL: Acta Tropica 53 (1): p59-72 1993
ISSN: 0001-706X
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Leishmania donovani promastigote glycoconjugate ligands, studied in our laboratory, that interact with the internalization receptors on BALB/c macrophages: the 'fucose mannose ligand' (FML), the 'phosphate mannogalactan ligand' (PMGL), and the 'lipopeptidephosphoglycan' (LPPD), interfered also with interaction between amastigotes and host cells in vitro. Among the three compounds studied, the FML was shown to be the most potent inhibitor of both promastigote and amastigote internalization, and to be present on parasite surface during the vertebrate-host cycle. The FML, but not the other two glycoconjugates, is a potent immunogen in rabbits (ELISA, agglutination and immuno-blots). Rabbit hyperimmune sera recognized essentially the 36 kDa band of FML. Mouse monoclonal antibodies against FML recognized either the 36 kDa or the 55 kDa band. No cross-reactivity between these two FML components was detected.

No antigenic similarity could be detected between the 36 and 59 kDa bands of FML and the 'GP63' (promastigote surface proteinase) major surface leishmanial antigen. The 36 kDa-glycoprotein was identified as the major FML antigenic fraction and designated 'GP36'. The integrity of the glycidic moiety was

necessary for its antigenicity. This *L. donovani* surface glycoprotein is apparently one of the major molecules involved in interactions between the parasite and the vertebrate host.

10/7/14 (Item 14 from file: 5)
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11409769 BIOSIS NO.: 199294111610
IDENTIFICATION OF A GP63 SURFACE GLYCOPROTEIN IN LEISHMANIA
-TARENTOLAE
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JOURNAL: FEMS Microbiology Letters 96 (1): p89-92 1992
ISSN: 0378-1097
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The promastigote stage of most if not all *Leishmania* species possesses an abundant surface glycoprotein of 63 kDa (gp63) that has protease activity. We show that the lizard parasite *Leishmania tarentolae* appears to lack the surface protease activity. *L. tarentolae* does, however, possess an approximately 63-kDa molecule that is antigenically cross-reactive with the *L. major* gp63. Additionally, the genome of *L. tarentolae* contains sequences that hybridise at high stringency to a *L. major* gp63 gene probe.

10/7/15 (Item 15 from file: 5)
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11174942 BIOSIS NO.: 199293017833
STRUCTURE AND ANTIGENICITY OF THE LIPOPHOSPHOGLYCAN FROM
LEISHMANIA-MAJOR AMASTIGOTES
AUTHOR: MOODY S F (Reprint); HANDMAN E; BACIC A
AUTHOR ADDRESS: WALTER AND ELIZA HALL INST MED RES, IMMUNOPARASITOL
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JOURNAL: Glycobiology 1 (4): p419-424 1991
ISSN: 0959-6658
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The lipophosphoglycan (LPG) of the intracellular amastigote form

of the protozoan parasite Leishmania major is chemically distinct from the LPG on the surface of the extracellular promastigote form. Amastigote LPG is composed of the monosaccharides galactose, glucose, mannose, glucosamine and inositol in the molar ratio 51:30:24:1:1; arabinose is absent. The lipid anchor comprises four alkylglycerols, with alkyl chain lengths 24:0, 22:0, 20:0 and 26:0 in the molar ratio 68:18:8:6. Phosphate is present at 4% w/w of total carbohydrate. HPLC gel permeation reveals LPG to be a polydisperse family of molecules Mr 100-6 kDa. The results from immunological studies with LPG-directed antibodies are consistent with amastigote LPG having the expected tripartite structure of GPI-anchor, a core glycan and the phosphorylated disaccharide repeat backbone. Human sera from L. major patients bound amastigote LPG in enzyme-linked immunosorbent assays.

10/7/16 (Item 16 from file: 5)
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10315683 BIOSIS NO.: 199090100162
TRYPANOSOMA-CRUZI DIFFERENTIAL EXPRESSION AND DISTRIBUTION OF AN 85-KDA POLYPEPTIDE EPITOPE BY IN-VITRO DEVELOPMENTAL STAGES
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LOYENS M; TAIBI A; MARTY B; VELGE P; RIZVI F; CAPRON A
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JOURNAL: Experimental Parasitology 71 (2): p207-217 1990
ISSN: 0014-4894
DOCUMENT TYPE: Article
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LANGUAGE: ENGLISH

ABSTRACT: The expression by Trypanosoma cruzi developmental stages of an 85-kDa polypeptide epitope defined by the 155D3 monoclonal antibody (mAb) has been investigated. Immunoprecipitation revealed the presence of an 85-kDa antigen in the NP-40 soluble extract of parasites freshly released from infected fibroblasts; this antigen was not found in epimastigote and Leishmania infantum promastigote. Indirect immunofluorescence revealed that the mAb 155D3 failed to react with trypomastigotes, whereas extracellular amastigotes were heavily stained. Positive organisms displayed either surface or polar

fluorescence. Since the same mAb immunoprecipitated the 85-kDa antigen in both radioactive iodine- and methionine-labeled trypomastigote detergent soluble extracts, the reactive epitope is likely to be hidden in a cryptic site in trypomastigotes. An alternative explanation for the negative immunofluorescence on trypomastigotes and the positive immunoprecipitation is the presence, in the extracts, of a small population of parasites already expressing the 155D3 epitope. Immunoelectron microscopy revealed that the target epitope is heterogeneously distributed among the populations of differentiating parasites. Two types of immunogold labeling were observed: (a) mAb revealed a high amount of reactive material associated with the periphery of the parasites and (b) a label was observed on the inner surface of peripheral vacuoles that might correspond to cross sections of inflated flagellar pockets and in association with vesicles which were released by the parasites. The surface expression of the epitope recognized by the 155D3 mAb was followed by fluorescence-activated cell-sorting analysis. The results showed that the epitope is increasingly accessible during trypomastigote differentiation *in vitro*. Taken together, these results suggest that the epitope reacting with the 155D3 mAb is heavily expressed on extracellular amastigotes after the transformation process and, thus, appears to be developmentally regulated.

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10187195 BIOSIS NO.: 199089105086
IDENTIFICATION OF CYTOPLASMIC SOLUBLE ANTIGENS RELATED TO THE MAJOR SURFACE ANTIGENS OF LEISHMANIA
-BRAZILIENSIS-BRAZILIENSIS AND LEISHMANIA-DONOVANI-CHAGASI
AUTHOR: KUTNER S (Reprint); PELLERIN P; BRENIERE S F
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JOURNAL: Parasitology Research 76 (3): p185-191 1990
ISSN: 0932-0113
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: This study describes the identification of aqueous-soluble antigens in Leishmania promastigotes immunologically and

biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis* *braziliensis* and *L. donovani* *chagasi* promastigotes, extracted and separated by partitioning

in the detergent Triton X-114, were analyzed. Immunoblotting of the extracted proteins, using homologous antisera, showed recognition of a

72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-kDa surface antigen of *L. d. chagasi*. The respective homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigens determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic

and the respective hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that could be directly attributed to the major *L. b.*

braziliensis and *L. d. chagasi* antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

10/7/18 (Item 18 from file: 5)
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10187143 BIOSIS NO.: 199089105034
THE MAJOR SURFACE GLYCOPROTEIN GP63 IS PRESENT IN BOTH LIFE STAGES OF LEISHMANIA

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JOURNAL: Molecular and Biochemical Parasitology 38 (1): p25-32 1990

ISSN: 0166-6851

DOCUMENT TYPE: Article

RECORD TYPE: Abstract

LANGUAGE: ENGLISH

ABSTRACT: Leishmania exist as extracellular promastigotes which multiply in the gut of the sandfly insect vector and as intracellular amastigotes which divide in the phagolysosome of mononuclear phagocytic

cells of the mammalian host. Promastigotes express a major surface glycoprotein of 63 kDa, referred to as GP63. The expression of GP63 in both Leishmania life stages was studied using rabbit antibodies

against native GP63 as well as rabbit antibodies against recombinant GP63 that was synthesized in an Escherichia coli expression system. Immunofluorescence staining detected GP63 in intracellular amastigotes contained within a macrophage cell line and within freshly isolated lesion amastigotes. Western blot analysis using anti-recombinant GP63 antibodies also demonstrated that amastigotes synthesize GP63 which may undergo differential post-translational processing as compared to promastigote GP63.

10/7/19 (Item 19 from file: 5)
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10165613 BIOSIS NO.: 199089083504
LEISHMANIA-TROPICA CHARACTERIZATION OF A LIPOPHOSPHOGLYCAN-LIKE ANTIGEN RECOGNIZED BY SPECIES-SPECIFIC MONOCLONAL ANTIBODIES
AUTHOR: JAFFE C L (Reprint); PEREZ M L; SARFSTEIN R
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JOURNAL: Experimental Parasitology 70 (1): p12-24 1990
ISSN: 0014-4894
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Species-specific monoclonal antibodies to Leishmania tropica. T11 and T13-15, recognize membranal and secreted antigens. The membrane form of the antigen migrates on sodium dodecyl sulfate-polyacrylamide gels with a diffuse molecular weight from 15 to 50 kDa and can be labeled with palmitic acid, myoinositol, galactose, glucosamine, and inorganic phosphate. Both phosphate and sugarlabeled material were isolated from metabolically labeled promastigotes by affinity chromatography on antibodies coupled to Sepharose 4B. No binding to Ricinus communis agglutinin was observed. This material behaves like lipophosphoglycans from other Leishmania but contains unique species-specific epitopes. It is susceptible to cleavage by phospholipase C and after digestion no longer partition as into the detergent phase following a Triton X-114 extraction. All four monoclonal antibodies appear to recognize carbohydrate epitope on the lipophosphoglycan since

periodate treatment of this material bound to nitrocellulose essentially eliminated antibody binding. In addition, T15 binding could be blocked by 5 mM mannose-6-P04 and fructose-1- or 6-P04, but not by mannose, glucose, fructose, or the additional P04 derivatives examined. The antibodies recognize a similar but not identical epitope, as demonstrated by a competitive radioimmunoassay using 125I-labeled T11, T13, and T15. Expression of surface antigen is elevated during the promastigote stationary phase.

10/7/20 (Item 20 from file: 5)
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09175995 BIOSIS NO.: 198886015916
THE MAJOR SURFACE PROTEIN OF LEISHMANIA PROMASTIGOTES IS A
FIBRONECTIN-LIKE MOLECULE
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JOURNAL: European Journal of Immunology 18 (3): p473-476 1988
ISSN: 0014-2980
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: The major surface glycoprotein of Leishmania chagasi promastigotes showed cross-reactivity with fibronectin (Fn), a large glycoprotein that is a major constituent of the extracellular matrix of most mononuclear cells. Polyclonal and monoclonal antibodies against Fn precipitated two molecules of 63-58 kDa from the lysates of both 125I and [35S]methionine-labeled promastigotes. In addition, a monoclonal antibody against a 15-kDa fragment of Fn containing the Arg-Gly-Asp-Ser (RGDS) sequence and several polyclonal monospecific mouse antibodies against a synthetic RGDS peptide also recognized the above two molecules. The attachment of Leishmania promastigotes to mouse peritoneal macrophages *in vitro* was partially inhibited when promastigotes were treated with F(ab')2 fragment of an anti-Fn IgG. Identical results were obtained by saturating the Fn receptors on macrophages using different peptides containing the RGDS sequence. Moreover, antigen preparations rich in glycoprotein 63 could efficiently promote the attachment and spreading of 3T3 mouse fibroblasts

to surfaces coated with the antigen. These results clearly suggest that the gp63 of *L. chagasi* promastigotes is an Fn-like molecule that shares certain biological and molecular characteristics with Fn.

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09175976 BIOSIS NO.: 198886015897
IDENTIFICATION OF MONOMERIC AND OLIGOMERIC FORMS OF A MAJOR
LEISHMANIA-INFANTUM ANTIGEN BY USING MONOCLONAL ANTIBODIES
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JOURNAL: Infection and Immunity 56 (5): p1180-1186 1988
ISSN: 0019-9567
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Ten monoclonal antibodies (MAbs) produced against isolated *Leishmania infantum* membranes were used as probes of *L. infantum* membrane antigens. Western blots of *L. infantum* membranes, sodium dodecyl sulfate solubilized and heated at 100°C before analysis of sodium dodecyl sulfate-polyacrylamide gel electrophoresis, showed that all 10 MAbs recognized a band at 58 kilodaltons (kDa). However, when solubilized membranes were not heated, 2 of the 10 MAbs recognized, in addition to the 58-kDa band, bands of higher molecular weight. Limited digestion of heated or nonheated membranes showed that both groups of MAbs (i.e., not capable or capable of binding to the high-molecular-weight bands) recognized the same proteolytic digests. Hydrophilic forms of the above proteins, possessing proteolytic activity, were detected and isolated by gel filtration. Protein staining of the isolated monomer analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis, under reducing and heating conditions, revealed incomplete reduction of the 58-kDa protein. The reduced form of the 58-kDa protein migrated at 63 to 65 kDa and was not recognized by the MAbs. These results suggest the existence of a monomeric and an oligomeric form of the 58-kDa antigen. The observed inhibition of *Leishmania* promastigote-macrophage binding caused

by MAbs representative of the two groups (capable of oligomeric and/or monomeric antigen recognition) suggest that the 5 8-kDa monomer and oligomer play an important role in promastigote-macrophage interaction.

We suggest that the 58-kDa *L. infantum* antigen is the major surface Leishmania antigen (p63) identified by others.

10/7/22 (Item 22 from file: 5)
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08682162 BIOSIS NO.: 198784036311
CLONING OF A GENE ENCODING THE IMMUNODOMINANT SURFACE ANTIGEN
OF LEISHMANIA-DONOVANI PROMASTIGOTES
AUTHOR: HEATH S (Reprint); CHANCE M L; HOMMEL M; CRAMPTON J M
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JOURNAL: Molecular and Biochemical Parasitology 23 (3): p211-222 1987
ISSN: 0166-6851
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: This study describes the characterisation of externally oriented surface peptides of both morphological forms of *Leishmania donovani*, the causative agent of visceral leishmaniasis (kala-azar). Using ^{125}I surface labelling techniques and peptide extraction in the detergents Triton X-100 and Triton X-114, a major iodinable promastigote peptide at 63 kDa or 65 kDa (depending on detergent used) was identified. This peptide was demonstrated to be the immunodominant membrane peptide of *L. donovani* and was strongly recognised by human sera from parasitologically confirmed cases of kala-azar. This peptide was not demonstrated on the surface of tissue amastigotes, although *in vitro* translations of poly(A⁺) RNA from both promastigotes and amastigotes demonstrated that both forms possessed mRNA that directs the synthesis of a 63 kDa peptide. It is suggested therefore that in amastigotes this peptide may be a processed antigen. We also report the isolation of a recombinant cDNA clone in the bacteriophage vector λ gt10 which encodes a 63 kDa polypeptide that is recognised by human kala-azar sera. It is proposed that this surface peptide could be used in a specific immunodiagnostic test for leishmaniasis.

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08148191 BIOSIS NO.: 198681112082

EXPRESSION AND SIZE HETEROGENEITY OF A 63-KILODALTON MEMBRANE
GLYCOPROTEIN

DURING GROWTH AND TRANSFORMATION OF LEISHMANIA-MEXICANA-
AMAZONENSIS

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JOURNAL: Molecular and Biochemical Parasitology 18 (2): p197-210 1986

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DOCUMENT TYPE: Article

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LANGUAGE: ENGLISH

ABSTRACT: Our previous work by immunoprecipitation with a specific monoclonal antibody showed multiple, closely apposed electrophoretic bands of a major surface antigen specific to the promastigote stage of Leishmania mexicana amazonensis.

Here, we analyzed the antigen during growth and transformation of this parasite with particular emphasis on the origin of the multiple bands. Immunobinding assays revealed the presence of the antigen throughout all phases of growth of cloned and uncloned promastigotes in

various media for different number of generations. More antigen is expressed by promastigotes grown in Medium 199 plus fetal bovine serum

than those in serum-supplemented Schneider's medium or a defined medium;

however, this is clone-dependent. Purified monoclonal antibody coupled to

Affi-Gel 10 gave a high capacity of antigen binding, resolving four electrophoretic bands of 60-66 kDa. A 63 kDa membrane protein, representing one of the four bands, become predominant after

[³⁵S]methionine label and chase. Pretreatment of promastigotes with 10

μg ml⁻¹ tunicamycin reduces the antigen to a single band of 54 kDa. Treatment of the antigen bound to the affinity gel with endoglycosidase-H produces similar, but less complete effect. These results indicate glycosylation of this antigen with asparagine-linked oligosaccharides, which appears to account at least in

part for its expression as multiple, closely apposed bands during biosynthesis. Binding of fluorescein isothiocyanate-labeled 6H12 monoclonal IgG or Fab to the promastigotes showed an even distribution of

the antigen over the cell surface and its capping upon the addition of rabbit anti-mouse IgG. Additional hybridomas prepared against

amastigotes yielded monoclonal antibodies which recognized surface antigens common to both stages of the parasite.

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07219928 BIOSIS NO.: 198477051839
RADIO IODINATION AND IDENTIFICATION OF EXTERNALLY DISPOSED MEMBRANE
COMPONENTS OF LEISHMANIA-TROPICA
AUTHOR: GARDINER P R (Reprint); DWYER D M
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JOURNAL: Molecular and Biochemical Parasitology 8 (4): p283-296 1983
ISSN: 0166-6851
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: ENGLISH

ABSTRACT: Two methods, one involving a lactoperoxidase-glucose oxidase coupled reaction and the other employing the insoluble catalyst 1,3,4,6-tetrachloro-3 α ,6- α -diphenyl-glycoluril (Iodo-Gen), were used to label the surface membrane of promastigotes of *L. t. major*. Both methods labeled .apprx. 20 proteins or glycoproteins (apparent size range 10-110 kDa [kilodaltons]) in a qualitatively similar manner; however, the lactoperoxidase method labeled 1 additional constituent (260 kDa). By omission of both enzymes, or of Iodo-Gen, by comparison of radioactivity incorporated by particulate and soluble cell fractions, and through the action of proteases on live, labeled promastigotes, the surface-labeling specificity of both procedures was confirmed. Immunoprecipitation of Triton X-100 extracts of labeled cells with rabbit antisera revealed a minimum of 12 (7 major) protein antigens in the homologous system and different but cross-reactive protein species from 2 other isolates of *L. tropica*. Lectin precipitation of radiolabeled surface components was possible with concanavalin A (but not with other lectins tested) identifying a minimum of 12 glycoproteins. Two of these glycoproteins (120 and 88 kDa) were not recognized by rabbit antiserum.

10/7/25 (Item 1 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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11168063 Genuine Article#: 617TU Number of References: 64
Title: Characterization of *Leishmania donovani* antigens
encapsulated in liposomes that induce protective immunity in
BALB/c

mice

Author: Afrin F; Rajesh R; Anam K; Gopinath M; Pal S; Ali N (REPRINT)
Corporate Source: Indian Inst Chem Biol, Infect Dis Grp, 4 Raja S-C
Mullick

Rd/Calcutta 700032/W Bengal/India/ (REPRINT); Indian Inst Chem
Biol, Infect Dis Grp, Calcutta 700032/W Bengal/India/
Journal: INFECTION AND IMMUNITY, 2002, V70, N12 (DEC), P6697-6706
ISSN: 0019-9567 Publication date: 20021200
Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC
20036-2904

USA

Language: English Document Type: ARTICLE

Abstract: Leishmania donovani promastigote membrane

antigens (LAg) encapsulated in positively charged liposomes have been found to induce very significant levels of protection against experimental visceral leishmaniasis. The protectively immunized animals

exhibited profound delayed-type hypersensitivity and antibody responses. The extent of protection induced by the same antigens, however, varied depending on the charge of the vesicles, with maximum

induction by positively charged liposomes, followed by neutral liposomes and last negatively charged liposomes. Characterization of

LAgs and LAg entrapped in liposomes of different charges by Western blot

analysis revealed the immunodominance of gp63 in all three vaccine preparations. The strong reactivity of antigens in a restricted antigen profile that included, in addition to gp63, 72-, 52-, 48-, 45-, 39-, and 20-kDa components in neutral and positively charged liposomes contrasted with the reactivity of a greater number of

LAgs components in negatively charged liposomes. Resistance to visceral

leishmaniasis appears to depend on the immunity induced by gp63 and a

few select antigens in association with the right liposomes. A striking similarity between the immunogenic profile of partially purified soluble antigens and that of LAg in neutral and positively charged liposomes suggests the potentiality of these antigens in future vaccine studies of *L. donovani*.

10/7/26 (Item 2 from file: 34)

DIALOG(R) File 34:SciSearch(R) Cited Ref Sci
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08139365 Genuine Article#: 250FY Number of References: 61
Title: Molecular cloning and characterization of a novel repeat-containing

Leishmania major gene, ppg1, that encodes a membrane-associated form of proteophosphoglycan with a putative

glycosylphosphatidylinositol anchor
Author: Ilg T (REPRINT) ; Montgomery J; Stierhof YD; Handman E
Corporate Source: MAX PLANCK INST BIOLAB MEMBRANBIOCHEM, CORRENNSSTR
38/D-72076 TUBINGEN//GERMANY/ (REPRINT); ROYAL MELBOURNE
HOSP,WALTER &
ELIZA HALL INST MED RES/MELBOURNE/VIC 3050/AUSTRALIA/
Journal: JOURNAL OF BIOLOGICAL CHEMISTRY, 1999, V274, N44 (OCT 29), P
31410-31420
ISSN: 0021-9258 Publication date: 19991029
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE
PIKE, BETHESDA, MD 20814
Language: English Document Type: ARTICLE
Abstract: Leishmania parasites secrete a variety of proteins that are
modified by phosphoglycan chains structurally similar to those of
the
cell surface glycolipid lipophosphoglycan. These proteins are
collectively called proteophosphoglycans. We report here the
cloning
and sequencing of a novel Leishmania major proteophosphoglycan
gene, ppg1. It encodes a large polypeptide of approximately 2300
amino
acids. The N-terminal domain of approximately 70 kDa exhibits 11
imperfect amino acid repeats that show some homology to
promastigote surface glycoproteins of the psa2/gp46
complex. The large central domain apparently consists exclusively
of
approximately 100 repetitive peptides of the sequence
APSASSSSA(P/S)SSSSS(+/-S). Gene fusion experiments demonstrate
that
these peptide repeats are the targets of phosphoglycosylation in
Leishmania and that they form extended filamentous structures
reminiscent of mammalian mucins. The C-terminal domain contains a
functional glycosylphosphatidylinositol anchor addition signal
sequence, which confers cell surface localization to a normally
secreted Leishmania acid phosphatase when fused to its C
terminus. Antibody binding studies show that the ppg1 gene
product is
phosphoglycosylated by phosphoglycan repeats and cap
oligosaccharides.
In contrast to previously characterized proteophosphoglycans, the
ppg1
gene product is predominantly membrane-associated and it is
expressed
on the promastigote cell surface. Therefore this
membrane-bound proteophosphoglycan may be important for direct
host-parasite interactions.

06844481 Genuine Article#: ZW149 Number of References: 58
Title: Human and murine immune responses to a novel *Leishmania* major recombinant protein encoded by members of a multicopy gene family
Author: Webb JR; CamposNeto A; Ovendale PJ; Martin TI; Stromberg EJ; Badaro
R; Reed SG (REPRINT)
Corporate Source: INFECT DIS RES INST, 1124 COLUMBIA ST, SUITE 200/SEATTLE//WA/98104 (REPRINT); INFECT DIS RES INST, /SEATTLE//WA/98104 ; CORIXA CORP, /SEATTLE//WA/98104; UNIV WASHINGTON, DEPT PATHOBIOL/SEATTLE//WA/98195; UNIV FED BAHIA,/SALVADOR/BA/BRAZIL/
Journal: INFECTION AND IMMUNITY, 1998, V66, N7 (JUL), P3279-3289
ISSN: 0019-9567 Publication date: 19980700
Publisher: AMER SOC MICROBIOLOGY, 1325 MASSACHUSETTS AVENUE, NW, WASHINGTON, DC 20005-4171
Language: English Document Type: ARTICLE
Abstract: Vaccination of BALB/c mice with *Leishmania* major promastigote culture filtrate proteins plus *Corynebacterium parvum* confers resistance to infection with *L. major*. To define immunogenic components of this protein mixture, we used sera from vaccinated mice to screen an *L. major* amastigote cDNA expression library. One of the immunoreactive clones thus obtained encoded a novel protein of *L. major* with a molecular mass of 22.1 kDa. The predicted amino acid sequence of this clone exhibited significant homology to eukaryotic thiol-specific-antioxidant (TSA) proteins. Therefore, we have designated this protein *L. major* TSA protein. Southern blot hybridization analyses indicate that there are multiple copies of the TSA gene in all species of *Leishmania* analyzed. Northern blot analyses demonstrated that the TSA gene is constitutively expressed in *L. major* promastigotes and amastigotes. Recombinant TSA protein containing an amino-terminal six-histidine tag was expressed in *Escherichia coli* with the pET17b system and was purified to homogeneity by affinity chromatography. Immunization of BALB/c mice with recombinant TSA protein resulted in the development of strong cellular immune responses and conferred protective immune responses against infection with *L. major* when the protein was combined with interleukin 12. In addition, recombinant TSA protein elicited in vitro proliferative responses from peripheral blood mononuclear cells of human leishmaniasis patients and significant TSA protein-specific antibody titers were detected in sera of both cutaneous-leishmaniasis and visceral-leishmaniasis patients. Together, these data suggest that the TSA protein may be useful as a component of a subunit vaccine.

against leishmaniasis.

10/7/28 (Item 4 from file: 34)
DIALOG(R) File 34:SciSearch(R) Cited Ref Sci
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06806832 Genuine Article#: ZT861 Number of References: 31
Title: Leishmania: Amastigotes synthesize conserved secretory acid phosphatases during human infection
Author: Ellis SL; Shakarian AM; Dwyer DM (REPRINT)
Corporate Source: NIAID,DIV INTRAMURAL RES, PARASIT DIS LAB, CELL BIOL SECT, NIH, 9000 ROCKVILLE PI/BETHESDA//MD/20892 (REPRINT); NIAID,DIV INTRAMURAL RES, PARASIT DIS LAB, CELL BIOL SECT, NIH/BETHESDA//MD/20892
Journal: EXPERIMENTAL PARASITOLOGY, 1998, V89, N2 (JUN), P161-168
ISSN: 0014-4894 Publication date: 19980600
Publisher: ACADEMIC PRESS INC JNL-COMP SUBSCRIPTIONS, 525 B ST, STE 1900,
SAN DIEGO, CA 92101-4495
Language: English Document Type: ARTICLE
Abstract: Leishmania donovani is the major causative agent of Old World human visceral leishmaniasis (VL). In vitro, both promastigotes and axenic amastigotes of *L. donovani* constitutively secrete soluble acid phosphatases (SAcPs), which contain conserved antigenic epitopes. These SAcPs are the most abundant and best characterized secretory proteins of this parasite. The aim of this study was to determine whether this enzyme was produced by intracellular amastigotes during the course of human infection. To that end, sera from acutely infected leishmaniasis patients were tested for anti-SAcP antibodies using *L. donovani* promastigote culture supernatants. Our results showed that VL patient sera from different endemic foci immunoprecipitated parasite SAcP enzyme activity. Further, these VL patient sera recognized the 110- and 130-kDa SAcPs in both Western blots and radioimmunoprecipitation assays. Results of tunicamycin experiments demonstrated that VL patient anti-SAcP antibodies were directed against the polypeptide backbone of the parasite SAcPs. In addition, both radiolabeled *L. donovani* SAcPs and native enzyme activities were immunoprecipitated by sera from patients with various forms of cutaneous leishmaniasis. Together these studies demonstrate that Leishmania amastigotes produce SAcPs during the course of human infections.

10/7/29 (Item 5 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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03622572 Genuine Article#: PT102 Number of References: 48
Title: IDENTIFICATION OF A 94-KILODALTON ANTIGEN ON LEISHMANIA
PROMASTIGOTE FORMS AND ITS SPECIFIC RECOGNITION IN HUMAN AND
CANINE VISCERAL LEISHMANIASIS

Author: ROLAND L; ZILBERFARB V; FURTADO A; GENTILINI M
Corporate Source: 1 RUE LAMARTINE/F-38000 GRENOBLE//FRANCE//; UNIV
PARIS

06,PARASITOL LAB/F-75643 PARIS 13//FRANCE//; INST PASTEUR,UNITE
IMMUNOPARASITOL/F-75724 PARIS//FRANCE//; FDN OSWALDO CRUZ,CTR
PESQUISAS

AGGEU MAGALHAES/RECIFE/PE/BRAZIL/

Journal: PARASITE IMMUNOLOGY, 1994, V16, N11 (NOV), P599-608

ISSN: 0141-9838

Language: ENGLISH Document Type: ARTICLE

Abstract: We have analysed by immunoblotting sera from humans and
dogs with

visceral leishmaniasis, from the Old World as well as the New.

When

lysates of promastigotes are used as antigens, antibodies against
a 94 kDa Leishmania component are detected, regardless of
the age and geographical origin of the patient, the serum antibody
titre as measured by indirect immunofluorescence, and the number
of

arcs in counterimmunoelectrophoresis. Low dilutions of sera from
patients with Old and New World cutaneous leishmaniasis did not
react

with the 94-kDa antigen, whatever the species of
Leishmania used as antigens. Sera from patients with other
infections than leishmaniases, or without infection, are
negative, even

at low dilution. Anti-94 kDa antibodies were detected in the sera
of Leishmania-infected dogs from both the Old and the New World.
When lysates of leishmania mexicana axenic amastigotes are used
as antigens, the 94-kDa antigen was little or none
identified by sera from humans and dogs with visceral

leishmaniasis,
and never recognized by control sera. Thus, the specific
recognition of
the 94-kDa promastigote antigen in human and canine
visceral leishmaniasis suggests that this antigen could be a
potential candidate in the differential immunodiagnosis of the
disease.

10/7/30 (Item 6 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

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03393010 Genuine Article#: PB879 Number of References: 57
Title: CHARACTERIZATION OF PHOSPHOGLYCAN-CONTAINING SECRETORY
PRODUCTS OF
LEISHMANIA
Author: ILG T; STIERHOF YD; WIESE M; MCCONVILLE MJ; OVERATH P
Corporate Source: MAX PLANCK INST BIOL, MEMBRANBIOCHEM ABT, CORRENSSTR
38/D-72076 TUBINGEN//GERMANY/; MAX PLANCK INST BIOL, MEMBRANBIOCHEM
ABT/D-72076 TUBINGEN//GERMANY/; UNIV DUNDEE, DEPT BIOCHEM/DUNDEE DD1
4HN//SCOTLAND/
Journal: PARASITOLOGY, 1994, V108, S, PS63-S71
ISSN: 0031-1820
Language: ENGLISH Document Type: ARTICLE
Abstract: This article presents an overview on
phosphoglycan-containing
components secreted by the insect and mammalian stages of several
species of Leishmania, the causative agents of leishmaniasis in
the Old and New World. Firstly, promastigotes of all three species
considered, L. mexicana, L. donovani and L. major, shed
lipophosphoglycan (LPG) into the culture medium possibly by
release of
micelles from the cell surface. Like the cell-associated LPG,
culture supernatant LPG is amphiphilic and composed of a
lysoalkylphosphatidylinositol-phosphosaccharide core connected to
species-specific phosphosaccharide repeats and oligosaccharide
caps.
Secondly, all three species release hydrophilic phosphoglycan.
Thirdly,
all three species appear to secrete proteins covalently modified
by
phosphosaccharide repeats and oligosaccharide caps. In the case of
promastigotes of L. mexicana, these components are organized as
two
filamentous polymers released from the flagellar pocket: the
secreted
acid phosphatase (sAP) composed of a 100 kDa phosphoglycoprotein
and a protein-containing high-molecular-weight-phosphoglycan
(proteo-HMWPG) and fibrous networks likewise composed of
phosphoglycan
possibly linked to protein. Structural analyses and gene cloning
suggest that the parasites can covalently modify protein regions
rich
in serine and threonine residues by the attachment of
phosphosaccharide
repeats capped by oligosaccharides. We propose that the networks
formed
in vitro correspond to fibrous material previously demonstrated
in the
digestive tract of infected sandflies. In the case of L.
donovani, the
sAP is also modified by phosphoglycans but contains neither

proteo-HMWPG nor does it aggregate to filaments. Finally, *L. mexicana* amastigotes release proteo-HMWPG via the flagellar pocket into the parasitophorous vacuole of infected macrophages. This material appears to be released into the tissue of the infected mammal upon rupture of infected macrophages during lesion development. This secretory product may contribute to the pathology of lesion development.

10/7/31 (Item 7 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
(c) 2009 The Thomson Corp. All rts. reserv.

02868765 Genuine Article#: ML423 Number of References: 26
Title: CHARACTERIZATION OF A LEISHMANIA ANTIGEN ASSOCIATED WITH CYTOPLASMIC VESICLES RESEMBLING ENDOCYTOSOME-LIKE STRUCTURE
Author: YAHIAOUI B; LOYENS M; TAIBI A; SCHONECK R; DUBREMETZ JF; OUAISSI MA
Corporate Source: INST PASTEUR,CTR IMMUNOL & BIOL PARASITAIRE,TRYPANOSOMATIDS RES LAB,INSERM,U167/F-59019 LILLE//FRANCE/;
INST PASTEUR,CTR IMMUNOL & BIOL PARASITAIRE,TRYPANOSOMATIDS RES LAB,INSERM,U167/F-59019 LILLE//FRANCE/; INSERM,U42/F-59045 LILLE//FRANCE/
Journal: PARASITOLOGY, 1993, v107, DEC (DEC), P497-507
ISSN: 0031-1820
Language: ENGLISH Document Type: ARTICLE
Abstract: In the present study we have used antibodies to Leishmania major promastigote antigens which were eluted from a glutathione-agarose column (LmGbp) and could identify several parasite components among different Leishmania species by using immunoprecipitation and Western blot techniques. The results also showed that some of LmGbp are present among the molecules released into the culture medium. Moreover, immunofluorescence assays clearly demonstrated that LmGbp are expressed by intracellular amastigotes. The electron micrographs of thawed cryosections of *L. major*-infected cells revealed that the antigens were associated with the membrane of the phagocytic vacuole. Moreover, the Western blot technique allowed us to identify, using other Leishmania species extracts and anti-LmGbp antibodies, a major polypeptide of an apparent molecular mass of 66 kDa. Immunofluorescence studies suggested that the 66 kDa polypeptide is associated with intracytoplasmic vesicles. Cryosections of Leishmania promastigotes improved the fine

structure preservation of the organelles and enabled a number of features to be seen, particularly the structures considered as vesicles, which appeared as a complex tubulo-vesicular structure resembling mammalian cell endosomes and Leishmania organelles previously named 'megasomes'. Further studies using antibodies against the native 66 kDa protein will be needed to investigate the localization of the protein at the ultrastructural level and to follow its intracellular vesicular traffic.

10/7/32 (Item 8 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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02781521 Genuine Article#: MC858 Number of References: 63
Title: MONOCLONAL-ANTIBODIES DIRECTED AGAINST LEISHMANIA SECRETED ACID-PHOSPHATASE AND LIPOPHOSPHOGLYCAN - PARTIAL CHARACTERIZATION OF PRIVATE AND PUBLIC EPITOPEs
Author: ILG T; HARBECKE D; WIESE M; OVERATH P
Corporate Source: MAX PLANCK INST BIOL, CORRENSSTR 38/D-72076 TUBINGEN//GERMANY/
Journal: EUROPEAN JOURNAL OF BIOCHEMISTRY, 1993, V217, N2 (OCT 15), P 603-615
ISSN: 0014-2956
Language: ENGLISH Document Type: ARTICLE
Abstract: Leishmania promastigotes, the stage of the parasite characteristic for the sandfly vector, express an abundant glycoconjugate, called lipophosphoglycan, at their surface. Lipophosphoglycan consists of lysoalkyl-sn-glycerophosphoinositol linked to a phosphosaccharide core conserved in all species, which is connected to P04-6Galbeta1,4Manalpha1 repeats with species-specific substitutions at the Gal residue; the repeats are capped by conserved and species-specific oligosaccharides. Most Leishmania species also secrete an acid phosphatase, which, in Leishmania mexicana, is a filamentous complex composed of a phosphorylated glycoprotein and non-covalently associated proteo-(high-molecular-mass) phosphoglycan.

The secreted acid phosphatase complex was used as an antigen to derive a panel of monoclonal antibodies (mAbs). A total of 25 mAbs

(17 novel and 8 previously described) were tested by different techniques for their specificity against lipophosphoglycan and secreted acid phosphatase from several Leishmania species. This comparison

and the modification of the antigens by chemical or enzymic treatments allowed a classification of the mAbs into several groups.

First, from 25 mAbs examined, 22 recognize lipophosphoglycan and the enzyme complex of *L. mexicana*; only three are specific for secreted acid phosphatase. Two of the latter group are also directed against carbohydrate structures, whereas the third mAb recognizes the 100-kDa polypeptide of the complex. The secreted acid-phosphatase-specific class detects antigen in the flagellar pocket of promastigotes while all anti-lipophosphoglycan mAbs bind to the cell surface.

Second, all 15 anti-lipophosphoglycan mAbs investigated in detail appear to be directed against the phosphosaccharide repeats or the cap structure rather than the phosphosaccharide core. Two mAbs recognize terminal cap-structures containing Man₁α1,2Man residues. Four antibodies are specific for *L. mexicana* and are probably directed against P04-6[Glc_β1,3]Gal_β1,4Man₁α1 repeats while six mAbs react with the unmodified repeats. Two antibodies specific for *Leishmania major* recognize Gal_β1,3-substituted repeats unique for lipophosphoglycan from this species.

Analysis by immunoblotting indicates that the high-molecular-mass proteo-phosphoglycan of *L. mexicana* secreted acid phosphatase carries epitopes for all anti-lipophosphoglycan mAbs suggesting the presence of capped phosphosaccharide repeats while the enzymically active glycoprotein subunit is modified by caps but probably not by repeats.

In the case of *Leishmania donovani* secreted acid phosphatase, the enzymically active polypeptide may be directly modified by repeats.

The mAbs are used to characterize changes in lipophosphoglycan structure, which occur in culture during the transition of promastigotes from the logarithmic to the stationary growth phase. Furthermore, testing the mAbs against seven species demonstrates their potential for serotyping *Leishmania*.

10/7/33 (Item 9 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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01138614 Genuine Article#: FZ314 Number of References: 30
Title: BIOCHEMICAL-CHARACTERIZATION OF THE PROTECTIVE MEMBRANE
GLYCOPROTEIN
GP46/M-2 OF LEISHMANIA-AMAZONENSIS
Author: RIVAS L; KAHL L; MANSON K; MCMAHONPRATT D
Corporate Source: YALE UNIV,SCH MED,DEPT EPIDEMIOL & PUBL HLTH,POB
3333,60
COLL ST/NEW HAVEN//CT/06510; YALE UNIV,SCH MED,DEPT EPIDEMIOL &
PUBL
HLTH,POB 3333,60 COLL ST/NEW HAVEN//CT/06510
Journal: MOLECULAR AND BIOCHEMICAL PARASITOLOGY, 1991, V47, N2,
P235-243
Language: ENGLISH Document Type: ARTICLE
Abstract: Biochemical features of the immunologically protective,
membrane
glycoprotein GP46/M-2 of Leishmania amazonensis have been
investigated. The protein appears to have a single carbohydrate
side chain of approximately 3 kDa, representing 7% of the mass of the
mature GP46/M-2 protein. Experiments removing this carbohydrate
side chain from GP46/M-2 indicate that the carbohydrate is not
involved in
the epitope recognized by the monoclonal antibody, M-2. As this
monoclonal antibody recognizes a species-specific epitope, these
data suggest that this determinant is defined by the polypeptide
portion of
the molecule. Studies employing the VSG-lipase as well as
anti-CRD
antibody clearly indicate that the molecule is anchored to the
surface membrane of the promastigote via a
phosphatidylinositol-linked lipid anchor. Neither the
carbohydrate side chain nor the lipid anchor appear to be responsible for the
apparent refractoriness of this protein to protease digestion,
suggesting that properties of the polypeptide itself may be
responsible. These data are discussed in terms of recent
DNA-derived
protein sequence of the GP46/M-2.

10/7/34 (Item 1 from file: 50)
DIALOG(R)File 50:CAB Abstracts
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0008538736 CAB Accession Number: 20033189694
Immune complex antigens as a tool in serodiagnosis of kala-azar.

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Kalyani,
Kalyani 741 235, West Bengal, India.
Molecular and Cellular Biochemistry volume 253 (1/2): p.191-198
Publication Year: 2003
ISSN: 0300-8177
Digital Object Identifier: 10.1023/A:1026095328695
Additional Title: Special issue: recent advances in molecular
physiology
Publisher: Kluwer Academic Publishers Dordrecht, Netherlands
Language: English
Record Type: Abstract
Document Type: Journal article

The 63 kDa surface antigen of Leishmania promastigotes is one of the most important virulent factors in establishing the host parasite relationship. This glycoprotein is revealed by surface iodination study as well as by metabolic labeling and immunoblot methods. In search of this specific antigen for serodiagnosis, immune complexes (ICs) were isolated from kala-azar patient sera and analysed by SDS-PAGE and Western immunoblotting. The immunoblot of kala-azar IC with patient sera, anti-promastigote sera and anti gp63 sera detected the major antigen of 55 kDa. This recognition suggests that 55 kDa antigen and gp63 have common antigenic epitope(s). Normal IC did not react with anti gp63 sera indicating absence of this antigen in normal IC. To confirm the parasitic origin of the 55 kDa antigen of kala-azar IC, in vitro IC was formed with parasite antigen and acid dissociated kala-azar IC antibody. This indicated the antigenic similarity of the 55 kDa antigen and gp63 antigen of the parasite. This also suggested that the former antigen may have been processed from gp63. In summary, identification of parasite antigen (55 kDa) in IC of kala-azar patients' sera may be useful in developing a serodiagnostic assay of visceral leishmaniasis. Several other antigens are visualized in kala-azar IC when developed with patient sera. But specificity and efficacy of these antigens have not yet been evaluated in serodiagnosis of the disease.

42 reference

10/7/35 (Item 2 from file: 50)
DIALOG(R)File 50:CAB Abstracts
(c) 2009 CAB International. All rts. reserv.

0008526162 CAB Accession Number: 20033177260

Extracellular release of the surface metalloprotease, gp63, from Leishmania and insect trypanosomatids.

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Parasitology Research volume 91 (3): p.229-237

Publication Year: 2003

ISSN: 0932-0113

Digital Object Identifier: 10.1007/s00436-003-0960-0

Publisher: Springer-Verlag Berlin, Germany

Language: English

Record Type: Abstract

Document Type: Journal article

Protease activity was found in spent culture medium collected from

Leishmania donovani , L. mexicana , L. major , as well as the insect trypanosomatids, Crithidia luciliae and Leptomonas seymouri . Released

protease activity increased linearly over time and was correlated to

promastigote density. In SDS-PAGE, zymogram gels showed that the protease's molecular weight ranged from 43-100 kDa. Spent culture medium proteases were blocked by the metallo-protease inhibitors,

1,10-phenanthroline and Z-Tyr-Leu-NHOH, but not by bestatin, leupeptin,

ABESF, pepstatin A, E-64 or aprotinin. Monoclonal and/or polyclonal

antibodies to the leishmanial gp63 reacted with the released Crithidia ,

Leptomonas , L. major and L. donovani proteases. Cell surface biotinylation and immune precipitation using gp63-specific antibodies

showed that >34% of the released protease originated from the

surface. Antibodies against the Trypanosoma brucei variable surface glycoprotein cross-reactive determinant (CRD) did not recognize this activity, suggesting that the gp63 is not cleaved from the

cell surface by a parasite phospholipase, but is released by an alternative mechanism.

48 reference

10/7/36 (Item 3 from file: 50)
DIALOG(R)File 50:CAB Abstracts
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0008432384 CAB Accession Number: 20033079688

Characterization of Leishmania donovani antigens encapsulated in liposomes that induce protective immunity in BALB/c mice.

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Swati Pal; Nahid Ali

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Infectious Diseases Group, Indian Institute of Chemical Biology,
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S. C. Mullick Rd., Calcutta 700 032, India.

Infection and Immunity volume 70 (12): p.6697-6706

Publication Year: 2002

ISSN: 0019-9567

Digital Object Identifier: 10.1128/IAI.70.12.6697-6706.2002

Publisher: American Society for Microbiology (ASM) Washington,
USA

Language: English

Record Type: Abstract

Document Type: Journal article

Leishmania donovani promastigote membrane antigens (LAG) encapsulated in positively charged liposomes have been found to induce very significant levels of protection against experimental visceral leishmaniasis. The protectively immunized animals exhibited profound delayed-type hypersensitivity and antibody responses. The extent of protection induced by the same antigens, however, varied depending on the charge of the vesicles, with maximum induction by positively charged liposomes, followed by neutral liposomes and last negatively charged liposomes. Characterization of LAg and LAg entrapped in liposomes of different charges by Western blot analysis revealed the immunodominance of gp63 in all three vaccine preparations. The strong reactivity of antigens in a restricted antigen profile that included, in addition to gp63, 72-, 52-, 48-, 45-, 39-, and 20-kDa components in neutral and positively charged liposomes contrasted with the reactivity of a greater number of LAg components in negatively charged liposomes. Resistance to visceral leishmaniasis appears to depend on the immunity induced by gp63 and a few select antigens in association with the right liposomes. A striking similarity between the immunogenic profile of partially purified soluble antigens and that of LAg in neutral and

positively charged liposomes suggests the potentiality of these antigens in future vaccine studies of *L. donovani*.
64 reference

10/7/37 (Item 4 from file: 50)
DIALOG(R)File 50:CAB Abstracts
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0008119239 CAB Accession Number: 20013022440
Regulation of GP63 mRNA stability in promastigotes of virulent and attenuated *Leishmania chagasi*.

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Molecular and Biochemical Parasitology volume 112 (1): p.51-59
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Document Type: Journal article

GP63 is a 63-kDa glycoprotein that is abundantly expressed on the surface of all *Leishmania* species and is involved in several steps of promastigote infection of host cells. *Leishmania chagasi* [*Leishmania infantum chagasi*] has at least 18 haploid msp (major surface protease) genes encoding GP63 that are divided into three classes, msp S, msp L or msp C, according to their unique 3prime UTR sequences and differential expression. All three msp classes are constitutively transcribed during virulent promastigote growth in vitro, although msp L mRNA is most abundant during logarithmic phase and msp S mRNA predominates in stationary phase. Thus, the steady state levels of the msp L and msp S mRNAs are post-transcriptionally regulated. Using Actinomycin D to arrest transcription, we found that in virulent promastigotes the half-life ($t_{1/2}$) of msp L mRNA is coordinately modulated with growth phase, decreasing from a mean of 84 min. during early logarithmic growth to a mean of 17 min. at a stage intermediate between logarithmic and stationary phase. However, in attenuated

promastigotes, the t_{SUB} 1/2 of msp L RNA remains the same throughout parasite growth. In contrast to msp L RNA, the t_{SUB} 1/2 of msp S and msp C RNA is constant throughout all growth phases of both virulent and attenuated promastigote growth. The presence of the translation inhibitor cycloheximide increases the t_{SUB} 1/2 of msp L RNA 4-6-fold in both virulent and attenuated promastigotes at all growth phases. These results indicate that the t_{SUB} 1/2 of msp L RNA is maintained by at least two distinct mechanisms - one activated during growth to stationary phase and the other dependent on a labile negative regulatory protein factor(s).

36 reference

10/7/38 (Item 5 from file: 50)
DIALOG(R)File 50:CAB Abstracts
(c) 2009 CAB International. All rts. reserv.

0007947182 CAB Accession Number: 20000809086
Expression and immunological identification of recombinant GP63 of Leishmania major .
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Xinjiang Institute for Endemic Disease Control and Research,
Urumqi,
Xinjiang 830002, China.
Endemic Diseases Bulletin volume 15 (2): p.13-14, 16
Publication Year: 2000
ISSN: 1000-3711
Language: Chinese Summary Language: English
Record Type: Abstract
Document Type: Journal article

Recombinant Leishmania major surface glycoprotein GP63 was expressed in Escherichia coli XL1-Blue transformed with the recombinant plasmid PAS 26, which was constructed from the pBluescript M13 plasmid and the cloned GP63 gene from L. major . The molecular weight of the expression product was 55 kDa after dissociation from beta-galactosidase. A low titre of cross reaction was seen between immune sera against recombinant GP63 and beta-galactosidase from bacteria. A high titre in ELISA was obtained using recombinant GP63 with immune sera

against Leishmania promastigote antigen and with sera from kala-azar patients. It is concluded that this recombinant GP63 could be useful in serological diagnosis of kala-azar and research on immunization against the disease.

2 reference

10/7/39 (Item 6 from file: 50)
DIALOG(R)File 50:CAB Abstracts
(c) 2009 CAB International. All rts. reserv.

0007522001 CAB Accession Number: 19980802636
Cloning of the gp63 surface protease of Leishmania infantum . Differential post-translational modifications correlated with different infective forms.
Gonzalez-Aseguinolaza, G.; Almazan, F.; Rodriguez, J. F.; Marquet, A.; Larraga, V.
Centro de Investigaciones Biologicas, CSIC, Velazquez 144, Madrid, Spain.
Biochimica et Biophysica Acta, Molecular Basis of Disease volume 1361 (1) : p.92-102
Publication Year: 1997
ISSN: 0925-4439
Language: English
Record Type: Abstract
Document Type: Journal article

The gp63 gene from Leishmania infantum [L. infantum infantum] PB75 was cloned and characterized. The sequence analysis of the gene indicated the existence of a high degree of conservation with the other Old World species L. major and L. donovani . The similarity was lower with New World species with the exception of L. chagasi [L. infantum chagasi] which showed a strikingly high percentage of identity (99-100%). In L. infantum the gp63 gene expressed 2 polypeptides of 58 and 60 kDa , respectively, which showed a similar proteolytic activity. The 60 kDa polypeptide was expressed during the whole life cycle of the promastigote form of the parasite with a moderate increase at the stationary phase of growth, whereas the 58 kDa product, although slightly present in the logarithmic phase, notably increased its

expression during the highly infectious stationary phase. RNA analysis showed that the presence in *L. infantum chagasi* of these 2 polypeptides correlated with 2 RNA molecules and with the degree of parasite infectivity, whereas in the case of *L. infantum infantum* a single 3 kb messenger RNA was detected through the whole promastigote life cycle. The data indicated that in *L. infantum infantum*, the differences in gene expression of the gp63 protease family according to parasite phase of growth seems to be due to a differential pattern of glycosylation of the polypeptides which correlates with the different infective forms of the promastigote form of the parasite. Nucleotide sequence data have been submitted to GenBank under accession number U48798.

46 reference

10/7/40 (Item 7 from file: 50)
DIALOG(R)File 50:CAB Abstracts
(c) 2009 CAB International. All rts. reserv.

000622255 CAB Accession Number: 19902070150

Identification of cytoplasmic soluble antigens related to the major surface antigens of *Leishmania braziliensis braziliensis* and *L. donovani chagasi*.

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Parasitology Research volume 76 (3): p.185-191
Publication Year: 1990
ISSN: 0044-3255
Language: English
Record Type: Abstract
Document Type: Journal article

This study describes the identification of aqueous-soluble antigens in *Leishmania* promastigotes immunologically and biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis braziliensis* and *L. donovani chagasi* promastigotes, extracted and separated by partitioning in the detergent Triton X-114, were analyzed. Immunoblotting of the extracted proteins, using homologous antisera, showed recognition of a 72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-

kDa surface antigen of *L. d. chagasi*. The respective homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigenic determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted

with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic and the respective

hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that

could be directly attributed to the major *L. b. braziliensis* and *L. d.*

chagasi antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

This study describes the identification of aqueous-soluble

antigens in *Leishmania* promastigotes immunologically and biochemically closely related to the major surface antigen. Proteins from surface-iodinated *L. braziliensis* *braziliensis* and *L. donovani chagasi* promastigotes, extracted and separated by partitioning in

the detergent Triton X-114, were analyzed. Immunoblotting of the extracted

proteins, using homologous antisera, showed recognition of a 72-kDa labeled, amphiphilic antigen of *L. b. braziliensis* and a 65-kDa surface antigen of *L. d. chagasi*. The respective homologous sera also recognized non-labeled hydrophilic antigens, similar in their apparent molecular weights to the major surface antigens. The amphiphilic and hydrophilic antigens of each species were found to share common antigenic determinants, inasmuch as monospecific antibodies that recognized the amphiphilic protein reacted

with the hydrophilic antigen. Structural homology was also obtained in the peptide-digestion profiles of the amphiphilic and the respective

hydrophilic major antigens. Zymogram assay showed that both amphiphilic and hydrophilic fractions displayed proteolytic activity that

could be directly attributed to the major *L. b. braziliensis* and *L. d.*

chagasi antigens. The hydrophilic antigens found in this study are probably not hydrolytic products of the surface antigens and occur in large quantities in the promastigote cytosol.

24 reference

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0003195596 SUPPLIER NUMBER: 1999195672
Interaction of Leishmania gp63 with cellular receptors for
fibronectin
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Journal: Infection and Immunity (Infect. Immun.), v67, n9,
(4477-4484),
1999, United States
PUBLICATION DATE: September 7, 1999 (19990907)
CODEN: INFIB
ISSN: 0019-9567 eISSN: 2009-003X
RECORD TYPE: Abstract; New
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 37

The most abundant protein on the surface of the promastigote form of the protozoan parasites *Leishmania* spp. is a 63-kDa molecule, designated gp63 or leishmanolysin. Because gp63 has been shown to possess fibronectin-like properties, we examined the interaction of gp63 with the cellular receptors for fibronectin. We measured the direct binding of *Leishmania* to human macrophages or to transfected mammalian cells expressing human fibronectin receptors. *Leishmania* expressing gp63 exhibited modest but reproducible adhesion to human macrophages and to transfected CHO cells expressing alpha4/beta1 fibronectin receptors. In both cases, this interaction depended on gp63 but occurred independently of the SRYD sequence of gp63, because parasites expressing gp63 with a mutated SRYD sequence bound to macrophages and alpha4/beta1 receptor-expressing cells as well as did wild-type parasites. The contribution of gp63 to parasite adhesion was more pronounced when the assays were performed in the presence of complement, suggesting that the receptors for complement and fibronectin may cooperate to mediate the efficient adhesion of parasites to macrophages. The interaction of gp63 with fibronectin receptors may also play an important role in parasite internalization by macrophages.

Erythrocytes to which gp63 was cross-linked were efficiently phagocytized by macrophages, whereas control erythrocytes opsonized with complement alone bound to macrophages but remained peripherally attached to the outside of the cell. Similarly, parasites expressing wild-type gp63 were rapidly and efficiently phagocytized by resting macrophages, whereas parasites lacking gp63 were internalized more slowly. This rapid internalization of gp63-expressing parasites was dependent on the betal integrins, because pretreatment of macrophages with monoclonal antibodies to the betal integrins decreased the internalization of gp63-expressing parasites. These observations indicate that complement receptors are the primary mediators of parasite adhesion; however, maximal parasite adhesion and internalization may require the participation of the betal integrins, which recognize fibronectin-like molecules such as gp63 on the surface of the parasite.

10/7/42 (Item 1 from file: 72)
DIALOG(R) File 72:EMBASE
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0079685472 EMBASE No: 2003394515
Identification of a gene in *Leishmania infantum* encoding a protein that contains a SP-RING/MIZ zinc finger domain
Garcia-Estrada C.; Reguera R.M.; Villa H.; Requena J.M.; Muller S.; Perez-Pertejo Y.; Balana-Fouce R.; Ordonez D.
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Biochimica et Biophysica Acta - Gene Structure and Expression (Biochim. Biophys. Acta Gene Struct. Expr.) (Netherlands) October 1, 2003, 1629/1-3 (44-52)
CODEN: BBGSD ISSN: 0167-4781
DOI: 10.1016/j.bbbaexp.2003.07.001
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English
NUMBER OF REFERENCES: 34
The SP-RING or Miz zinc finger domain that is related to the classical

RING-finger motif, defines a class of proteins that can act as E3-like factors in the pathway of small ubiquitin-related modifier (SUMO) conjugation. This family includes the mammalian protein inhibitor of activated STAT (PIAS) proteins and related proteins from lower eukaryotes.

Here we report the existence of a gene in *Leishmania infantum*, present as two identical copies placed upstream of each MAT2 gene copy, and

transcribed as a single (similar) 2.2 kb mRNA both in the logarithmic and

stationary phases of the promastigote stage. This gene encodes a 47 kDa protein that has been named LORIEN. LORIEN is circumscribed to the cell periphery and it is antigenic during *L. infantum* infection of dogs and hamsters. Strikingly, this novel protein contains a highly conserved SP-RING/Miz zinc finger domain, raising the possibility that a

SUMO or ubiquitin-like system may exist in this microorganism. (c) 2003

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10/7/43 (Item 2 from file: 72)
DIALOG(R)File 72:EMBASE
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0075659276 EMBASE No: 1994087105
Leishmania major parasites share an epitope with the murine CD3-T cell receptor complex
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European Journal of Immunology (EUR. J. IMMUNOL.) (Germany)
March 1,
1994, 24/3 (503-507)
CODEN: EJIMA ISSN: 0014-2980
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English
NUMBER OF REFERENCES: 20

After immunization of BALB/c mice with a low molecular mass fraction (FrD; <= 31 kDa) isolated from a soluble extract of *Leishmania* major promastigotes, a panel of monoclonal antibodies (mAb) was obtained.

One of these antibodies (mAb 9C) recognized a cytosol-associated antigen from *L. major* of approximately 21 kDa as shown by

Western blot and immunoprecipitation. In addition, mAb 9C reacted with surface structures of murine splenic T cells and T cell clones. Reactivity was confined to murine cells, but was not strain restricted.

Immunoprecipitation studies and surface-labeling experiments with CD4 SUP + T cell clones and the T cell receptor (TCR) SUP -CD3 SUP - T cell line TG40 transfected with V alpha/beta chains from human TCR and concomitant co-expression of murine CD3 suggested that mAb 9C binds to an epitope located within the murine CD3-TCR complex. In addition, mAb 9C induced strong T cell proliferation. We conclude that *L. major* parasites share an epitope with the murine CD3-TCR complex which is functionally important for T cell activation.

10/7/44 (Item 1 from file: 73)
DIALOG(R)File 73:EMBASE
(c) 2009 Elsevier B.V. All rts. reserv.

0074694298 EMBASE No: 1991199109
Metacyclogenesis of *Leishmania* spp: Species-specific in vitro transformation, complement resistance, and cell surface carbohydrate and protein profiles
Bandyopadhyay P.; Ghosh D.K.; De A.; Ghosh K.N.; Chaudhuri P.P.; Das P.; Bhattacharya A.
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Journal of Parasitology (J. PARASITOL.) (United States) July 24,
1991
' 77/3 (411-416)
CODEN: JOPAA ISSN: 0022-3395
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

Metacyclic (stationary) and logarithmic (log) forms of promastigotes of *Leishmania donovani* and *Leishmania major* were characterized in several ways. The highly active metacyclic forms were larger with more protein and less carbohydrate. The flagellum increased in length 2.4 times in *L. major* as compared to 1.8 times in *L. donovani*. Resistance to

complement-mediated lysis by normal human serum of in vitro grown Leishmania promastigotes was related to the species, the growth phase in culture, and also the temperature. Metacyclic forms of both species had a much increased resistance to killing by normal serum at different temperatures. Differences in membrane-exposed carbohydrates were detected by fluorescein-conjugated lectins. Peanut agglutinin and Ulex agglutinin I differentiated log and stationary phase promastigotes of *L. major*. Higher amounts of acid phosphatase were demonstrated in the metacyclic phase. Differences in polypeptides were detected by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Two polypeptides of approximately 51 and 114 kDa were found exclusively in metacyclic promastigotes of both species, whereas 38- and 23-kDa polypeptides were lost or reduced during transformation from log to metacyclic phase promastigotes of *L. donovani*. In addition, a 75-kDa polypeptide was expressed only in metacyclic promastigotes of *L. major*.

10/7/45 (Item 2 from file: 73)
DIALOG(R)File 73:EMBASE
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0074667156 EMBASE No: 1991171967
Localization of the plasma membrane and mitochondrial H SUP +-ATPases in
Leishmania donovani promastigotes
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European Journal of Cell Biology (EUR. J. CELL BIOL.) (Germany)
July
1, 1991, 54/1 (95-101)
CODEN: EJCBD ISSN: 0171-9335
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

Immunochemical methods were used to characterize the proton-translocating ATPases (H SUP +-ATPases) of the plasma membrane and mitochondrion of *Leishmania donovani* promastigotes. Antisera directed against the plasma membrane H SUP +-ATPase of *Saccharomyces cerevisiae* reacted with a 66 kDa membrane protein of *L. donovani* promastigotes. By immunocytochemistry, the antiserum was shown to label the cell and

flagellar surface of promastigotes as well as the Golgi apparatus and the membrane of intracellular organelles. The target antigen was shown to possess ATPase activity resembling the leishmanial H SUP +-ATPase activity. Antisera raised against the beta-subunit of the F SUB OF SUB 1-ATPase of Escherichia coli reacted with a 56 kDa protein in L. donovani promastigotes. Ultrastructurally, the anti-beta-subunit antibody was exclusively associated with the mitochondrion in these cells. This antiserum immunoprecipitates ATP hydrolytic activity typical of the F SUB 1 beta-subunit activity of the mitochondria of higher eukaryotes.

10/7/46 (Item 3 from file: 73)
DIALOG(R)File 73:EMBASE
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0074598336 EMBASE No: 1991103836
Antigenic specificity of the 72-kilodalton major surface glycoprotein of Leishmania braziliensis braziliensis
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Journal of Clinical Microbiology (J. CLIN. MICROBIOL.) (United States)
April 11, 1991, 29/3 (595-599)
CODEN: JCMID ISSN: 0095-1137
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

We examined the expression and the antigenicity of the major surface polypeptides of Leishmania braziliensis braziliensis and Leishmania donovani chagasi, parasites which commonly coexist in the same endemic areas of Bolivia. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis protein profiles from surface-iodinated promastigotes showed the presence of a unique iodinatable polypeptide of 72 kDa on the L. b. braziliensis surface and of two major components of 65 and 50 kDa exposed at the surface of L. d. chagasi. Comparison of the peptide digestion profiles of the major iodinated polypeptides of both strains showed no similarity between the maps of the 72- and the 65-kDa polypeptides of L. b. braziliensis and L. d. chagasi, respectively. Immunoprecipitation of surface-labeled L. b. braziliensis Nonidet P-40 extracts with 35 serum specimens obtained

from Bolivian patients with cutaneous and mucocutaneous leishmaniasis showed that all serum specimens recognized predominantly the 72 kDa antigen and high-molecular-mass proteins in some cases. The recognition patterns were independent of the geographical origin of the patient, the type of lesion, and the serum antibody titer. Serum specimens from children with visceral leishmaniasis did not precipitate the L. b. braziliensis 72-kDa antigen. Hamster hyperimmune serum against L. b. braziliensis also recognized the 72-kDa surface antigen. However, this recognition was inhibited in the presence of the homologous nonlabeled antigen but not in the presence of heterologous (L. d. chagasi and *Trypanosoma cruzi*) antigens. The specific recognition of the 72-kDa surface antigen in both natural and experimental L. b. braziliensis infections suggests that this antigen could be a good candidate for use in the differential immunodiagnosis and prognosis of the disease.

10/7/47 (Item 4 from file: 73)
DIALOG(R)File 73:EMBASE
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0074554918 EMBASE No: 1991060418
Evidence of transferrin binding sites on the surface of
Leishmania promastigotes
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Journal of Biological Chemistry (J. BIOL. CHEM.) (United States)
December 1, 1990, 265/36 (22380-22385)
CODEN: JCCHA ISSN: 0021-9258
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

A glycoprotein of 78,000 molecular mass (78 kDa), associated with the membrane of *Leishmania infantum* promastigotes, was identified and immunopurified by monoclonal antibody (mAb) LD9 produced against isolated membrane preparations. mAb LD9 was subsequently found to bind to human transferrin, also of 78 kDa. Binding of LD9 to transferrin was completely abolished when the mAb was preabsorbed by *Leishmania* membranes, thereby indicating that the 78-kDa *Leishmania* membrane-associated glycoprotein and transferrin have common

antigenic epitope(s). The 78-kDa Leishmania membrane-associated protein was released in soluble nonaggregated form by mild treatment with acetic acid saline. Anti-transferrin polyclonal antibodies, recognized both the membrane-associated and the soluble form of the 78-kDa glycoprotein. The 78-kDa soluble form was characterized further as an iron-containing protein. The above data combined with iron uptake by promastigotes as demonstrated by the Prussian blue reaction indicate that the 78-kDa Leishmania membrane-associated glycoprotein is transferrin. The binding of SUP 125I-human transferrin to Leishmania-purified membrane preparations was then investigated. The results indicate the presence of a high affinity saturable binding site $[K(d) = 2.2 \text{ } 10^{-8} \text{ M}]$ that is specific for transferrin. We suggest that the 78-kDa glycoprotein recognized by mAb LD9 is transferrin that binds to the surface of Leishmania promastigotes via a transferrin receptor.

10/7/48 (Item 5 from file: 73)
DIALOG(R)File 73:EMBASE
(c) 2009 Elsevier B.V. All rts. reserv.

0074263395 EMBASE No: 1990157408
Biochemical evidence of the antigenic cell surface heterogeneity of Leishmania mexicana
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Parasitology Research (PARASITOL. RES.) (Germany) June 20, 1990,
76/4
(301-305)
CODEN: PARRE ISSN: 0044-3255
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

In the present study, an enzymatical and structural analysis of Leishmania mexicana cell-surface components was carried out, demonstrating that protease and acid phosphatase activities were present at the L. mexicana cell surface. These findings correlate with the expression of the main components detected on the surface of L. mexicana promastigotes: the 50-kDa component is responsible for the

acid phosphatase activity, whereas glycoprotein 65 (gp65) was characterized as the structural polypeptide of the surface protease. Furthermore, the 50- and 65-kDa antigens were found to be structurally different, inasmuch as no homology was observed in their peptide digestion profiles. The results presented in this communication confirm heterogeneity in the expression of the surface components of *L. mexicana* promastigotes at both the structural and the biochemical level.

10/7/49 (Item 6 from file: 73)
DIALOG(R)File 73:EMBASE
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0073714390 EMBASE No: 1988175283
The major concanavalin A-binding surface glycoprotein of *Leishmania donovani chagasi* promastigotes is involved in attachment to human macrophages
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Journal of Immunology (J. IMMUNOL.) (United States) August 15, 1988,
141/1 (265-272)
CODEN: JOIMA ISSN: 0022-1767
DOCUMENT TYPE: Journal; Article RECORD TYPE: Abstract
LANGUAGE: English SUMMARY LANGUAGE: English

Leishmania donovani, the protozoan causing visceral leishmaniasis, is an obligate intracellular parasite of mammalian macrophages. Considerable evidence has suggested that the ingestion of *L. donovani* promastigotes by macrophages occurs via receptors on the surface of the phagocyte. During this study, a glycoconjugate that may be involved in the receptor-mediated ingestion of *L. donovani chagasi* promastigotes was isolated from the parasite membrane. Octyl glucoside-soluble extracts of promastigote membranes contained a predominant doublet migrating at 60 kDa, seen by SDS-PAGE. The 60-kDa molecule was the major externally disposed promastigote surface protein labeled by ¹²⁵I, and it was the major Con A-binding protein on *L. donovani chagasi*, as

determined by Con A binding to parasite proteins transferred to nitrocellulose. Attachment of promastigotes to human monocyte-derived macrophages was inhibited by varying concentrations of the membrane extract containing both proteins, and adsorption of extracts on Con A-Sepharose resulted in both removal of the 60,000 M(r) glycoprotein and loss of the ability of extracts to inhibit promastigote attachment to human macrophages. After further purification of the 60-kDa glycoprotein by gel filtration, its inhibitory activity increased 45-fold over the unpurified membrane extract. Examination of Con A blots of stationary phase promastigotes isolated from an infected hamster revealed a marked loss in the major Con A-binding glycoprotein over 4 mo in *in vitro* culture after isolation from the rodent host, corresponding to a loss in infectivity of the promastigotes for hamsters. The results suggest that the major Con A-binding surface glycoprotein from *L. donovani chagasi* promastigotes is important in attachment to human macrophages, and may be a factor in parasite virulence for a mammalian host.

10/7/50 (Item 1 from file: 144)
DIALOG(R) File 144:Pascal
(c) 2009 INIST/CNRS. All rts. reserv.

08889818 PASCAL No.: 90-0057784
L'infectivite des promastigotes de *Leishmania* est associee a l'expression de leurs antigenes de surface
(Infectivity of *Leishmania* promastigotes is associated with surface antigenic expression)
RIZVI Farrukh; CAPRON Andre, Dir the
Univ.: Lille 1 Degree: Th. doct. : Immunol.
1989; 1989 145 p.
Availability: CNRS-T Bordereau
No. of Refs.: 165 ref.
Document Type: T (Thesis) ; M (Monographic)
Country of Publication: France
Language: French
Caracterisation des molecules de surface de *Leishmania chagasi* impliquees dans l'attachement du promastigote au macrophage.
L'infectivite des promastigotes est associee a l'expression d'une glycoproteine de surface de masse moleculaire 63 kDa (gp 63). mise en evidence d'une communauta antigenique entre la gp63 et la fibronectine. Il semble que le parasite puisse adhérer au macrophage grace aux recepteurs fibronectines de celui-ci

10/7/51 (Item 1 from file: 154)
DIALOG(R)File 154: MEDLINE(R)
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14317997 PMID: 11432790
Role of 67 kDa cell surface laminin binding protein of
Leishmania donovani in pathogenesis.
Bandyopadhyay K; Karmakar S; Ghosh A; Das P K
Molecular Cell Biology Laboratory, Indian Institute of Chemical
Biology,
Raja S.C. Mullick Road, Jadavpur, Calcutta 700 032, India.
Journal of biochemistry (Japan) Jul 2001, 130 (1) p141-8,
ISSN
0021-924X--Print Journal Code: 0376600
Publishing Model Print
Document type: Journal Article; Research Support, Non-U.S. Gov't
Languages: ENGLISH
Main Citation Owner: NLM
Record type: MEDLINE; Completed
The role that interaction with laminin may play in Leishmania
donovani infection was investigated. Binding of
(125)I-radiolabeled
laminin, in a liquid-phase assay, by the parasite was rapid,
saturable,
specific, reversible, and of high affinity. Using a Western
blotting
procedure, a 67 kDa laminin-binding protein (LBP) was identified from
the membrane of both the promastigote and amastigote forms of L.
donovani. Subsequently, the protein was purified by
affinity
chromatography. Immunofluorescence with a polyclonal antibody
against LBP
as well as flow cytometric analysis demonstrated its presence
at the
parasite surface. After stimulation with phorbol-12-myristate-13-acet
ate (PMA), U937 cells exhibited the ability to adhere to laminin
and LBP
specifically inhibited this adhesion. The reduced parasite adhesion
after
tunicamycin treatment suggested the importance of sugar residues
in cell
adhesion. Although co-administration of either laminin or LBP or
anti LBP
antibody reduced parasite virulence, resulting in a lower
level of
infection in the BALB/c mouse model, an in vitro
macrophage
culture-enhanced level of infection was observed in the
case of
laminin-coated parasites. The results collectively suggest a role
for LBP

in the interaction of the parasite with extracellular matrix elements, which may constitute a basis for the homing of the parasite to its physiological address.

Record Date Created: 20010702

Record Date Completed: 20011205

10/7/52 (Item 1 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

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08737821 PMID: 2450920

Monoclonal antibodies specific for the amastigote stage of Leishmania pifanoi. I. Characterization of antigens associated with stage- and species-specific determinants.

Pan A A; McMahon-Pratt D

Department of Epidemiology and Public Health, Yale University School of Medicine, New Haven, CT 06510.

Journal of immunology (Baltimore, Md. - 1950) (UNITED STATES)

Apr 1

1988, 140 (7) p2406-14, ISSN 0022-1767--Print Journal Code: 2985117R

Contract/Grant No.: AI-07136; AI; NIAID NIH HHS United States; AI-23004;

AI; NIAID NIH HHS United States

Publishing Model Print

Document type: Journal Article; Research Support, Non-U.S. Gov't;

Research Support, U.S. Gov't, P.H.S.

Languages: ENGLISH

Main Citation Owner: NLM

Record type: MEDLINE; Completed

Eight mAb were produced against membrane-enriched preparations of

Leishmania pifanoi amastigotes either grown in axenic culture (P-1 through P-6) or isolated from macrophage cell culture (P-7 and P-8). Two

mAb produced against promastigote membranes (P-9 and P-10) were found to be specific against this stage. Antibodies P-1 through P-8 on analysis

by radioimmune binding only reacted with determinants present on

amastigotes. mAb P-2, P-4, and P-8 also reacted with Leishmania amazonensis amastigotes but not promastigotes. No cross-reactions were found on any other species of Leishmania or with membranes of Trypanosoma cruzi epimastigotes or amastigotes. An indirect

immunofluorescence assay using mAb P-1 through P-8 confirmed the stage

specificity and binding to *L. pifanoi* axenically grown amastigotes, amastigotes within infected hamster tissue, and amastigotes within J774.1 macrophages. When Western blot analysis of amastigote membranes was conducted, one distinct group of molecules associated with *L. pifanoi*-specific determinants was identified. mAb P-1, P-3, P-5, P-7, and P-8 bound to molecules Mr 43 and 34 kDa. Promastigote-specific mAb P-9 recognized a diffuse pattern from 88 to greater than 200 kDa, and mAb P-10 localized a second class of proteins with Mr53 kDa. On immunoprecipitation of solubilized [³⁵S]methionine-labeled amastigotes, mAb P-2 recognized a doublet of Mr 35 and 33 kDa and another doublet at Mr 17.5 and 13.5 kDa. mAb P-4 and P-7 each precipitated a band at Mr 34 kDa. These studies indicate that antigenically the axenically cultured amastigote is closely related to macrophage-derived amastigote. These mAb and/or purified protein Ag may be useful in studying stage differentiation, monitoring transformation, and for further taxonomic, epidemiologic, and immunologic studies of New World leishmaniasis.

Record Date Created: 19880426

Record Date Completed: 19880426

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| S1 | 164 | AMAZONENSIS AND PROMASTIGOTE AND SURFACE AND ANTIGEN? |
| S2 | 51 | RD S1 (unique items) |
| S3 | 175 | AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR ESA) |
| S4 | 55 | RD S3 (unique items) |
| S5 | 177478 | ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR ESA) |
| S6 | 1112 | S5 AND (LEISHMANIA OR AMAZONENSIS) |
| S7 | 335 | RD S6 (unique items) |
| S8 | 298 | S7 NOT PY>2004 |
| S9 | 0 | S8 AND 42.5 |
| S10 | 52 | S8 AND KDA |

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| Set | Items | Description |
|-----|-------|---|
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| S2 | 51 | RD S1 (unique items) |
| S3 | 175 | AMAZONENSIS AND ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR |

PSA OR ESA)
S4 55 RD S3 (unique items)
S5 177478 ((PROMASTIGOTE AND SURFACE AND ANTIGEN?) OR PSA OR
ESA)
S6 1112 S5 AND (LEISHMANIA OR AMAZONENSIS)
S7 335 RD S6 (unique items)
S8 298 S7 NOT PY>2004
S9 0 S8 AND 42.5
S10 52 S8 AND KDA
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---Logging off of Dialog---

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